SEP 13 1927

VOL. 54 JUNE, 1927 No. 6

BULLETIN

OF THE

TORREY BOTANICAL CLUB

Editor

TRACY ELLIOT HAZEN

Associate Editors

Albert Francis Blakeslee
Cornelia Lee Carey
Alexander William Evans
Henry Allan Gleason
Alfred Gundersen
George Tracy Hastings
Marshall Avery Howe
Michael Levine
Sam F. Trelease
Arlow Burdette Stout

CONTENTS

Additions to the genus Rhododendron ..... WILLIAM TRELEASE 471

A study of seed development in three species of Erigeron with special reference to apogamy ..... CLINTON C. MCDONALD 479

The production of intumescences in Transparent apple by ethylene gas as affected by external and internal conditions
RAYMOND HAROLD WALLACE 489

INDEX TO AMERICAN BOTANICAL LITERATURE 545

PUBLISHED FOR THE CLUB

PRESS OF
THE NEW ERA PRINTING COMPANY
20 W. KING ST., LANCASTER, PA.

Entered at the Post Office at Lancaster, Pa., as second-class matter

The Intelligencer Printing Company

LANCASTER, PENNSYLVANIA

Printers and Binders of

Technical and Scientific

Books and Periodicals

Estimates Furnished

TORREYA

A bimonthly journal of botany

Articles on the local flora, non-technical accounts of common plants of all groups, botanical notes, book reviews, etc.

Published by the Torrey Botanical Club at $1.00 a year. Address the treasurer,

Mrs. Helen M. Trelease,
Box 49, Schermerhorn Hall, Columbia University, New York City

---

BIOLOGY SUPPLIES

Dissecting Instruments	Chemicals and Stains
Microscopes and Accessories	Microscope and Lantern Slides
Models and Charts	Naturalists' Supplies
Laboratory Glassware	Skeletons

Catalogues applying to any of these articles in which you are interested will be sent on request.

NEW YORK BIOLOGICAL SUPPLY CO.

General Supplies for the Biological Sciences

24 UNION SQUARE, NEW YORK CITY

Please mention the "Bulletin" when answering advertisements.

Additions to the genus Phoradendron

WILLIAM TRELEASE

In the decade since the publication of my monograph of this exclusively American genus[1] a number of current collections have been submitted to me for determination, and among them the following species have been recognized as new. Preoccupation with Piperaceae has so engrossed my attention during recent visits to the great herbaria that I have not examined collections which may have found their way into these herbaria directly, so that it is quite possible that added novelties of which I am ignorant may have been collected. Descriptions of the species here noted were given at the time to the botanists from whom specimens were received, so that it is possible that others than the three cited below may have been published already under the names here used; but if such publication has escaped my notice, no harm can come from repetition here.

1. **Phoradendron Goldmani** n. sp.

(BOREALES—FLAVESCENTES)

Rather large, not forked, the moderate branches without cataphyls, dioecious. Internodes rather short (2–5 × 20–50 mm.), at first papillate-puberulent like the very young foliage. Leaves elliptic- or obovate-oblong, very obtuse, about 1 × 3 cm., very shortly cuneate-subpetioled. Spikes solitary, elongated (90 mm. in fruit), papillate-puberulent, with some seven elongated joints annularly 6- to 12-flowered when pistillate; peduncle commonly of 2 joints 5 mm. long; scales truncate-connate, papillate and papillate-ciliate. Fruit apparently white, subglobose, glabrous, about 4 mm. in diameter, in widely separated whorls; sepals glabrous, closely inflexed.

Eastern Sierra Madre region of Mexico.—The type from Camargo, Tamaulipas (Goldman 748, 749, Jan. 1902).

2. **Phoradendron huricolum** n. sp.

(AEQUATORIALES—ANGUSTIFOLIAE)

Scarcely forked, the moderate branches with basal cataphyls only, dioecious. Internodes rather short (2–3 × 40–50 mm.),

[1] The Genus Phoradendron, Urbana, Illinois, The University of Illinois. 1916.

[The BULLETIN for May (**54**: 383–470) was issued 18 June 1927.]

smooth, from somewhat 2-edged becoming 4-sided. Cataphyls a single basal pair, short. Leaves oblong-lanceolate, obtuse, 1.3 × 8 cm., finely basinerved, cuneately subpetioled for about 5 mm. Spikes subsolitary, short (about 10 mm.), slender, smooth, with 2 or 3 oblong joints some 6-flowered in 4 + 2 series when pistillate; peduncle scarcely 2 mm. long; scales subtruncately connate. Fruit (immature) subglobose, 2 mm. in diameter, smooth; sepals closely inflexed.

Venezuelan region.—The type from Venezuela.

Specimens examined:—Venezuela. Vicinity of Cristobal Colon (Broadway 1778, on *Hura crepitans*,—the type at the New York Botanical Garden).

3. **Phoradendron pomasquianum** n. sp.

(AEQUATORIALES—ANGUSTIFOLIAE)

More or less forked or fastigiately branched, the slender and rather short branches with basal cataphyls only. Dioecious?. Internodes rather short (1–2, or below 5, × 25–40 mm.), smooth, terete or evanescently flattened. Cataphyls a single basal pair or followed at a small distance by a second pair, spreading and becoming white-margined. Leaves narrowly oblong, obtuse, scarcely 1 × 50 mm., finely basinerved, cuneately slender-petioled for scarcely 10 mm. Spikes solitary in the axils, rather short (scarcely 30 mm.), slender, smooth, with about 4 turbinate joints 12- to 20-flowered in 4 series when pistillate; peduncle about 5 mm. long. Fruit (immature) oblong, 2–3 × 4 mm., smooth; sepals inflexed.

Andean region.—The type from Ecuador.

Specimens examined:—Ecuador. High plains near Pomasqui at 2500 m. (Sodiro 22,—the type at the University of Illinois).

4. **Phoradendron pseudo-angustifolium** n. sp.

(AEQUATORIALES—ANGUSTIFOLIAE)

Closely resembling *P. angustifolium* in habit, measurements and details, except that the leaves are thicker and for this reason obscurely if at all nerved.

Andean region.—The type from Ecuador.

Specimens examined:—Ecuador, Las Juntas (Rose, Pachano and Rose 23212,—the type as sheet 1022732 in the U. S. National Herbarium).

5. **Phoradendron Roseanum** n. sp.

(AEQUATORIALES—ANDINAE)

Very large and coarse, varnished, scarcely forked, the long branches apparently with basal cataphyls only, probably

dioecious. Internodes rather long (6–8 × 70–100 mm. or more), smooth, ancipital or 2-winged becoming terete. Cataphyls?. Leaves falcately lanceolate, large (as much as 4 × 20 cm.), rather obtuse, about 7-nerved, cuneately wing-petioled for about 10 mm. Spikes commonly clustered, long (60–80 mm. in fruit), with about 4 turbinate joints some 40- to 50-flowered in 6 series; peduncle scarcely 5 mm. long; scales scarcely ciliate, subtruncate. Fruit (reddish?) globose, smooth, about 3 mm. in diameter; sepals closely inflexed.

Andean region.—The type from Ecuador.

Specimens examined:—Ecuador, Las Juntas (Rose, Pachano and Rose 23213, Sept. 28, 1918,—the type, as sheet 1022733, in the U. S. National Herbarium).

Marked by its glossy stem and infloresence and large elongated leaves: differing from other members of the group in its very long spikes with more numerous 6-ranked flowers.

6. **Phoradendron pifoense** n. sp.

(AEQUATORIALES—ANDINAE)

Scarcely forked, the rather long branches with basal cataphyls only, dioecious?. Internodes moderate (2–4 × 40–50 mm.), smooth, at first ancipitally flattened. Cataphyls basal, solitary. Leaves subfalcately lanceolate, obtuse, 2.5–3 × 8–9 cm., or becoming as much as 6 × 15 cm., rather sharply 7-nerved, cuneately subpetioled for scarcely 10 mm. Spikes more or less clustered in the axils, thick but only moderately (30–50 mm.) long, with about 4 oblong-obovoid joints about 20-flowered in 4 + 2 series when pistillate; peduncle scant 3 mm. long; scales somewhat ciliate-scarious-margined, subtruncate. Fruit red, dull, globose, 3–4 mm. in diameter; sepals inflexed.

Andean region.—The type from Ecuador.

Specimens examined:—Ecuador. Pifo (Mille 32,—the type at the University of Illinois); also reported for Chillos Valley at 2600 m.

7. **Phoradendron Pachanum** n. sp.

(AEQUATORIALES—ANDINAE)

More or less divaricately forking, the rather long branches with basal cataphyls only, androgynous. Internodes moderate or rather stout (2–4 × 25–50 mm.), smooth and drying glossy, little compressed. Cataphyls a basal pair, rounded. Leaves obliquely or dimidiately sublanceolate, obtuse, 2.5–5 × 9–14 cm., about 5-nerved, wing-petioled for 10 mm. Spikes clustered, rather short (scarcely 30 mm.), with about 4 oblong or slightly clavate joints some 24-flowered in 4 + 2, or 6, series,—

the uppermost short and few flowered; peduncle 2–4 mm. long. Fruit subglobose, smooth, 3–4 mm. in diameter; sepals closely inflexed.

Andean region.—The type from Ecuador.

Specimens examined:—Ecuador, Ambato, Tungurahua (A. Pachano 246,—the type in the U. S. National Herbarium).

8. **Phoradendron brachypodum** n. sp.

(AEQUATORIALES—POLYGYNAE)

With basal cataphyls only on the branches and androgynous?. Internodes rather long (3–4 × 40–50 mm.), smooth, the upper ancipitally compressed, scarcely dilated at the nodes. Cataphyls a single pair, closely basal, tubular. Leaves lance-oblong or oblanceolate, very obtuse, 1.5–2.5 × 5–8 cm., cuneately contracted for 5–10 mm. at base, dull, faintly subpinnately 5- or 7-nerved, spikes solitary in the axils, thick, apparently to become moderately (some 50 mm.) long, with 5 or 6 stout subclavate-oblong joints 60- to 80-flowered in about 6 series; peduncle 5 mm. long, the succeeding joint floriferous; scales tubular-subtruncate. Fruit?.

Andean region.—The type from Colombia.

Specimens examined:—Colombia. San Cristobal, near Usme (Brother Ariste-Joseph A 919,—the type).

9. **Phoradendron guascanum** n. sp.

(AEQUATORIALES—OBLIQUAE)

Large, brittle, with basal cataphyls only?, androgynous?. Internodes thick (5–10 mm.), granular-corrugated, terete or at first compressed, but little dilated at the nodes. Cataphyls apparently a nearly basal pair, followed at intervals of 1 or 2 cm. by 2 or 3 others, tubular. Leaves obliquely obovate or subdimidiately elliptic, very obtuse, becoming 10 × 13 cm., cuneately thick-petioled for about 10 mm., pinnately about 10-nerved with the midrib heavy and prominent, drying thick, hard and glossy. Spikes elongated?, with several subclavate thick joints, some 36-flowered in 6 series; peduncle?; scales tubular-subtruncate. Fruit?.

Andean region.—The type from Colombia.

Specimens examined:—Eastern paramos of Guasca, toward Gacheta (Brother Ariste-Joseph B 60, in 1921,—the type).

The obvious pinnate nerving of this species is in disaccord with the obscure (really palmate?) nerving of *P. obliquum*.

10. **Phoradendron Millspaughii** n. sp.

(AEQUATORIALES—QUADRANGULARES)

Scarcely forking, the moderate branches with basal cataphyls only, androgynous?. Internodes rather short (2 × 30–50 mm.), essentially smooth, quadrangular, somewhat compressed at the nodes. Cataphyls a single nearly basal pair, nearly separate, pale-margined. Leaves oblanceolate-oblong, submucronately obtuse; 1–1.5 × 4–6 cm., cuneately narrowed rather than petioled. Spikes solitary, moderate (30–50 mm.), with 3 or 4 elongated joints turbinately about 20-flowered in 4 or 4 + 2 series; peduncle scarcely 2 mm. long. Fruit (immature) ellipsoid, smooth, 3 mm. long: sepals closely inflexed.

Yucatan region.—The type from Yucatan.

Specimens examined:—Mexico. Forests of Suitun, Yucatan (Gaumer & Sons 23827, part 2, 1917,—the type, in the Herbarium of the Field Museum, as sheet 466268).

Closely related to *P. Gaumeri*, but distinct in its elongated leaves and more turbinately flowered spikes: and with foliage comparable with the Isthmian *P. ceibanum*, the Pacific *P. Townsendi*, and some forms of the Mexican *P. tamaulipense*.

11. PHORADENDRON RUSBYANUM Trel.

In Rusby, Mem. New York Bot. Garden **7**: 234. 1927.

(AEQUATORIALES—QUADRANGULARES)

Brazilian region.—The type from Bolivia.

Specimens examined:—Reyes, Bolivia (Rusby 1714,—the type).

12. PHORADENDRON CAERULESCENS Trel.

In Britton, Bull. Torr. Bot. Club **48**: 330. 1921.

(AEQUATORIALES—EMARGINATAE)

Venezuelan region.—The type from Trinidad.

Specimens examined:—Chacachacare Island, Trinidad (Britton and Hazen 1726,—the type); also from the same locality, on *Capparis* (Britton, Freeman and Watts, 2701, 2708).

13. PHORADENDRON CHAGUARAMASANUM Trel.

In Britton, Bull. Torr. Bot. Club **48**: 330. 1921.

(AEQUATORIALES—EMARGINATAE)

Venezuelan region.—The type from Trinidad.

Specimens examined:—Chaguaramas, Trinidad (Britton 2718,—the type).

14. **Phoradendron Baileyae** n. sp.

(AEQUATORIALES—LIGAE)

Somewhat pseudo-dichotomous, the moderately long branches with basal cataphyls only, glabrous, androgynous?. Internodes moderate or rather short (2–3 × 20–40 mm.), ancipitally or the uppermost slightly rhomboidally compressed. Cataphyls a single nearly basal pair. Leaves oblanceolate-oblong and narrow (scarcely 1 × 5 cm.) or the lower obovate-oblong and broader (1.5 cm.) rounded or submucronate at apex, cuneately narrowed to the scarcely petioled base, drying thick but rather evidently 5-nerved from the base. Spikes short (10–15 mm.), with about 3 rather slender nearly cylindric joints about 4-flowered near the middle; peduncle 2 mm. long, with subtruncately connate scales. Fruit yellow?, ellipsoid, 3 × 4 mm., smooth, with widely separated sepals.

Venezuelan region.—The type from the vicinity of Ciudad Bolivar, on the Orinoco (L. H. and Ethel Zoe Bailey 1401, Feb. 28, 1921).

15. **Phoradendron lapatanum** n. sp.

(AEQUATORIALES—HEXASTICHAE)

Scarcely forked, the rather long branches with basal catapyls only, androgynous?. Internodes moderate (3–6 × 30–70 mm.), somewhat microscopically papillate, ancipital or somewhat rhombic and dilated upwards, becoming terete. Cataphyls usually a single pair, but sometimes 2–3 pairs within less than 1 cm. of the base, subtruncately bifid, ciliate. Leaves round-ovate to obovate, very obtuse, narrowly revolute, 2–4 × 4–5 cm., drying dark and chartaceous with percurrent midrib, obscurely pinnately veined, petioled for about 10 mm. Spikes mostly solitary, rather long (20, becoming 40–50 mm.), with about 4 somewhat fusiform joints scarcely over 12-flowered in 6 series; peduncle scarcely 8 mm. long, sometimes with a basal pair of sterile or floriferous scales. Fruit apparently red, globose, slightly cellular-papillate, 4 mm. in diameter: sepals closely inflexed.

Antillean region.—The type from Cuba.

Specimens examined:—El Corte, Sta. Tomas, Peninsula of Lapata, Cuba (Roig & Cremata 2198, in 1920,—the type at the New York Botanical Garden).

Differing from *P. hexastichum* in its darkening smaller and often obovate leaves, and its larger differently colored fruit.

16. PHORADENDRON BARAHONAE Urb. & Trel.

In Fedde, Repert. 13: 444. 1914,—as Phoradendrum.

(AEQUATORIALES—PTERONEURAE)

Antillean region.—The type from Hispaniola.

Unfortunately the prior publication of this herbarium name was overlooked when Wright's herbarium name *P. cerinocarpum* was adopted in 1916.

17. **Phoradendron canzacotoi** n. sp.

(AEQUATORIALES—TUNAEFORMES)

Scarcely forking, the elongated branches with basal cataphyls only, androgynous?. Internodes rather short (5–10 × 20–40 mm.), thin-winged from a terete axis 3–5 mm. in diameter rather than flattened, slightly papillate, not striately ridged. Cataphyls a single short annular basal pair. Leaves minute, scale-like, marcescent. Spikes more or less clustered, very short (scarcely 5 mm.), of a single joint about 15-flowered in 4 series; peduncle very short. Fruit?.

Andean region.—The type from Ecuador.

Specimens examined:—Ecuador, Subtropical forest, Canzacoto, at 2,000 m. (Sodiro 14,—the type at the University of Illinois).

THE UNIVERSITY OF ILLINOIS,
URBANA, ILLINOIS.

A study of seed development in three species of Erigeron with special reference to apogamy

CLINTON C. McDONALD

(WITH PLATES 32, 33)

A study of the development of the embryo sac, fertilization, and seed formation in *Erigeron annuus* Pers., *Erigeron strigosus* Muhl., and *Erigeron Philadelphicus* L. was made in order to compare the species in regard to apogamy and double fertilization.

In 1900 Land (1900) gave an account of his observation of double fertilization in the two species—*E. strigosus* and *E. Philadelphicus*. With the exception of *Silphium laciniatum* L. (also observed by Land in 1900) the writer knows of no other report of double fertilization in the Compositae.

Masato Tahara (1921) recently reported the occurrence of apogamy in *E. annuus* which he collected in Japan.

Several plants belonging to genera within the Compositae are apogamous. Apogamy occurs in the genus *Antennaria* (Juel, 1898, 1900); *Chondrilla* (Rosenberg, 1912); *Erigeron* (Tahara, 1921); *Eupatorium* (Holmgren, 1916); *Hieracium* (Murbeck, 1904; Ostenfeld, 1904, 1906; Rosenberg, 1906, 1907); and *Taraxacum* (Raunkaier, 1903; Murbeck, 1904; Juel, 1904, 1905; Osawa, 1913; Sears, 1917; Stork, 1920).

The writer presents the two species *E. annuus* and *E. strigosus* as apogamous plants and *E. philadelphicus* as normal in seed development for this locality.

MATERIAL AND METHOD

The Erigerons, like other Compositae, are favorable for the study of the ovule and embryo sac, because many flowers are closely grouped into a single inflorescence. Therefore, when sections are made vertically through one of the heads, many ovules are cut at one time, and a single preparation will show many embryo sacs in slightly different stages of development. Only median longitudinal sections of the head give good median longitudinal sections of the ovule, but even then a single specimen will have fifty or sixty favorable sections of ovules for examination and comparison.

The material was collected in the vicinity of Bloomington, Indiana. The specimens of *E. annuus* and *E. strigosus* were gathered from the first to the twenty-first of June, while those for *E. Philadelphicus* were taken during the last half of May, the latter species flowering earlier.

Several collections were made for each plant at different localities and at different times of the day. Various ages of heads were obtained from the same plant in some cases, but duplicate ages from different plants were also examined in order to determine the uniformity of the material used. The lower part of the receptacle, the upper part of the involucre and the corolla tubes were cut away, leaving only enough of the receptacle to hold the ovaries in a normal position. All material was killed in the field, as soon as possible after removing the specimens. The aqueous solution of chrom-osmic-acetic acid (formula as published by Mottier) was used as a killing and fixing agent. At first the entire pruned head was plunged into the solution, but experience taught that the fixative affected the tissue better if the heads were divided longitudinally. After sectioning some of this material, however, it was found that the desirable ovaries in the center of the head were bent to one side as a result of the release of pressure so that median longitudinal sections could not be obtained for many embryo sacs. Finally the method of cutting away considerable portions of opposite sides of the head helped effect the penetration of the fixative and at the same time left the desirable ovaries through the center of the head in an excellent position for sectioning. Since material was abundant and rather easily prepared, new collections were made and handled by the improved method. The specimens were left in the fixing solution for twenty-four hours, and then washed thoroughly and dehydrated by passing through the different strengths of aqueous alcoholic solutions into absolute alcohol. Their next transfer was through two grades of alcoholic chloroform to pure chloroform, and finally they were embedded in paraffin for sectioning. Sections were cut from four to eight microns in thickness and stained with the triple stain of analin-safranin, gentian-violet, and orange G. This method as a whole gave unusually clear preparations for a detailed study.

DEVELOPMENT OF THE EMBRYO SAC OF ERIGERON ANNUUS

The sections of young heads which were still completely enclosed in the involucre showed the ovule in an early stage of its development. The young nucellus with a specially differentiated hypodermal cell was the most noticeable part of the structure (fig. 1). The beginning of the thick fleshy integument shows only as a mass of cells below and to the left, no growth up and around the nucellus having been made at this age of the ovule. This special differentiated cell would ordinarily be spoken of as the megaspore mother cell that is to give rise to the tetrad, but examination of many ovules showed that no tetrad is formed. It is then, only a morphological megaspore mother cell which functions as a megaspore and in a series of divisions develops into the embryo sac, as the following discussion and figures will show. This condition indicates the lack of the reduction division and suggests apogamy.

Figure 2 shows a section of the nucellus at an older stage of the ovule. Careful investigation proved this large prominent cell to be identical with the supposed megaspore mother cell of figure 1, and not the functional megaspore following tetrad divisions. Here the nucellus has grown in length and diameter, and the integument is developed as a thick fleshy mass of tissue which completely surrounds the nucellus and forms the micropyle. This growth comes about very quickly, and the megaspore mother cell with the nucleus in the resting stage has increased greatly in size. This growth seems to be due mainly to hydration, since the cytoplasm has become vacuolated. Some of the sections show large vacuoles, especially when the nucleus of the megaspore is late in dividing, as in figure 3.

The nucellus is always composed of a single layer of cells with dense cytoplasm and well defined nuclei. There is no distinct nutritive layer of tapetal-like tissue formed on the inner border of the thick fleshy integument. The layer of cells of this inner border, however, stain somewhat more densely than the general cells of the integument, especially in the older stages of development of the embryo sac. The nucellus has been included in these drawings, but none of the integument tissue is shown. The boundary lines for the figures represent only the inner border of the integument. The wall of the nucellus begins to disappear at

about the time that the original nucleus divides to form an embryo sac with two nuclei (fig. 4). Sometimes, however, the division is late and the condition will be as shown in figure 3. In either instance it is the upper end of the nucellus that is absorbed first, and the chalazal end may be seen as a cylinder of tissue in section standing up around the antipodal end of the embryo sac (figs. 3 and 5). In such cases there are always signs of further deterioration, and by the time the embryo sac is fairly well formed the nucellar tissue has all disappeared.

The continued rapid growth of the ovule is quite evident during the disappearance of the nucellus and is paralleled by an equally rapid development of the embryo sac. The nucleus of the megaspore mother cell (figs. 2 and 3) begins a series of divisions that result in the formation of the mature structure of figure 6. These divisions are in rapid succession: first, the two nuclei as in figure 4; then the four as in figure 5; and finally the seven cells of figure 6. The latter shows three antipodal cells, the three cells of the egg apparatus, and the two polar nuclei at the time of fusion to form the nucleus of the endosperm cell. The antipodal cells disorganize early, and it was hard to find a specimen to show them. Most of the sections of embryo sacs show no antipodal cells at this time. In the section shown in figure 6, there are three antipodals, but the writer has reason to believe that this number is not constant, since there were in some cases only two nuclei left in the lower end of the sac after the lower polar nucleus was gone. The early disorganization of the antipodal cells, however, would account for such a condition. The polar nuclei fuse early, and when the egg apparatus is in other details as represented in figure 6, the fusion is in the majority of cases already complete. These nuclei meet near the egg in a rather dense area of cytoplasm.

In speaking of the egg cell of the embryo sac the term will be used to mean the structure that is morphologically the egg but not in the sense of an egg as a gamete. The apogamous condition with the accompanying lack of a reduction division makes it necessary to look upon the egg as a cell with the number of chromosomes of the sporophyte. The cell is outstanding in that it is decidedly pear-shaped, with a long attenuated end extending into the micropyle. The nucleus is large and lies in the lower end of the cell in a dense mass of cytoplasm. The beaked end of the cell is invariably occupied by a large vacuole.

The synergids are more slender beaked cells, but somewhat pear-shaped. The beaked ends, which generally contain cytoplasm, extend into the micropyle along with the end of the egg cell. There is a large vacuole at the lower end of each cell, and sometimes smaller ones are distributed throughout the cytoplasm. The latter is never abundant in the synergid cells. The nuclei are small and are located, as a rule, near the large vacuole. These cells also disorganize early but not so soon as the antipodals. However, in sections of slightly older stages, which show the egg cell as has been described, there are no synergid cells.

Observation on many ovules failed to show anything that suggested fertilization. Since the question of apogamy was under consideration, a diligent study was made of stages at the time that fertilization would take place to be sure that no irregularity occurred.

At about the time that the synergids disappear, the endosperm nucleus, which has been in the resting stage, divides with the spindle at right angles to the long axis of the sac. Other divisions, which may be in any plane, follow in rapid succession until there is an endosperm built up around the egg cell and young embryo. At first free nuclei are present, but after the endosperm formation is well under way the cellular type is predominant.

Before there is much endosperm formed, the egg cell divides to make a few celled embryo. The first few divisions show the cell plates put down at right angles to the length of the embryo sac, and the resulting cells are sometimes binucleate. The nuclei themselves are generally binucleolate (fig. 7). Subsequent divisions may occur in any plane until there is a large dicotyledonous embryo that occupies the entire ovule. All the endosperm that was formed and the tissue of the integument are used in the building up of the embryo. When the latter is fully formed, the crushed out cells of the integument show as a thin membrane next to the wall of the achene.

EXPERIMENTAL EVIDENCE OF APOGAMY IN ERIGERON ANNUUS

It has been stated that the formation of the tetrad accompanied by the reduction division does not come about in the develop-

ment of the embryo sac of this plant. Also, nothing could be found with reference to fertilization. Both of these conditions indicate apogamy, and an experiment was performed in which all parts of the flower except the ovule were cut away in order to determine what development would be made under such a condition. This procedure removed the stigma and most of the style, as well as the stamens. The cut was made near the base of the corolla tube. Unless the pollen would germinate on the cut ends of the style, fertilization would be impossible. In this work the latter condition was assumed impossible, so that subsequent development was to be considered from the morphological egg itself. It was thought that the ovule would live long enough to show if the embryo sacs continued their development and if embryos were formed in them.

The material on which this work was done and the technique of the experiment need some further explanation before continuing with the results. Heads were selected in which the flowers were not yet open. From the experience gained in collecting separate stages of the specimens for the first part of this work, it is known that heads were selected in which the embryo sacs were not mature. With a little experience it became easy to cut away the parts of the flower at the base of the corolla tube. A very sharp safety razor blade proved to be a good tool for such work.

Many heads were decapitated at one time, and frequent visits were made to cut away any part of the flower that seemed to have regenerated. There is a marked tendency, it seems, for the cut ends to grow out again, possibly a tendency to regenerate the missing part. However, after a day or two of sunshine, the exposed ends of the stylar tissue became dry and demanded no further attention in this respect.

Decapitated heads were fixed daily after the second day of the experiment, and since in due time there was no outward appearance of any cessation in the development of the ovules, some of the heads were left in order to see if the achenes would ripen. The growth continued until the seeds were mature. A collection was made at the time that they were ready to fall away from the receptacle, and upon examination they seemed normal in outward appearance. Examination of the content of the achenes revealed well formed embryos which later upon

planting were found to be viable. There are always some ovules that never mature seed, and this is true for the three species from both the experimental and the normal condition.

Sections of the fixed material showed normal development of the later stages of the embryo sac and also of the embryo. So the experimental evidence confirmed the morphological study of the development of the embryo. This condition is the same as reported by Masato Tahara (1921) for *E. annuus* in Japan. He describes and figures thirteen chromosomes in the nucleus of the pollen grain; but twenty-six for the morphological egg cell or any somatic cell. No tetrad is formed according to his work. However, he uses the word parthenogenesis where in this paper the term apogamy is used. The morphological egg undoubtedly contains the double number of chromosomes and as a reproductive structure stands in the same relation to the sporophyte as the spore does to the gametophyte. Then, since there is no true functional sex cell to be considered here, a term that signifies the absence of gametes where they are normally encountered is preferable.

EVIDENCE OF APOGAMY IN ERIGERON STRIGOSUS

The same method was used in the investigation of this species as has been described for *E. annuus*. This is one of the plants in which Land (1900) reported double fertilization, but no evidence secured in the present study substantiates such a conclusion. Sections of the preserved material showed a surprising similarity between the two species. The size and appearance of the ovule; time of development of the integument; lack of a nutritive layer on the inner border of the integument; character and time of disappearance of the nucellus; and absence of a tetrad stage in the development of the embryo sac—all were as has been described for the preceding plant.

Also, the experiment upon the living plant in which the ovules were isolated from the other parts of the flower by removing the stamens and stigmas proved that viable seeds were set as was to be expected. This then adds *E. strigosus* to the number of apogamous plants.

DEVELOPMENT OF THE EMBRYO SAC OF ERIGERON PHILADELPHICUS

Erigeron Philadelphicus does not conform to the description that has been given for the other two species. Consequently, it will be necessary to treat this species in a special development and give enough drawings accompanied by sufficient discussion to make the differences clear.

In the sections of very young ovules, the megaspore mother cell appears in the developing nucellus, which is not yet surrounded by the integument. In this respect this species is similar to *E. annuus* (fig. 1) and *E. strigosus*, but the subsequent behavior shows a great difference. The wall of the nucellus, like that of the other two species, is one layer of cells in thickness and grows to the usual size for the species, but at this stage it contains the tetrad of megaspores instead of the megaspore mother cell (fig. 8). In a single head of florets several stages like that of figure 8 may be found. They are all the more striking since this stage is lacking in the other two species. Another difference is that the integument shows the beginning of a distinct nourishing layer of cells, which is tapetal-like in character, on the inner border adjacent to the nucellus (fig. 8).

Slides showing the tetrads also show the lower megaspore consuming the other three cells, as well as the surrounding nucellus (fig. 9). It is to be noted that the latter begins to disappear at the chalazal end instead of at the micropylar end as was the case in the two preceding species. The functional megaspore, which is always the lower cell of the tetrad, becomes large and contains many vacuoles by the time the entire nucellus is consumed. This development of a tetrad indicates a reduction division and the formation of an embryo sac containing the typical egg cell, which is in reality a gamete of the gametophyte generation and not a diploid cell appearing morphologically as an egg.

There is nothing of importance in the development of the functional megaspore into the mature embryo sac. The ovule enlarges, the nutritive layer becomes very distinct down to the antipodal cell region, and soon the condition is as represented in figure 10. The egg cell is the typical pear-shaped structure with the well defined nucleus in the lower end and a vacuole just above

it. The upper end is elongated and seems to aid the beaked synergids to fill the opening into the micropyle. The latter are small and disappear at the time of fertilization. In one case two nuclei were present in one of the synergids cells. Like the egg, the synergids are not contained within a cell wall membrane.

The polar nuclei come together below the egg and fuse before fertilization. The resulting endosperm nucleus then takes a position near the egg cell and remains there until the egg and sperm unite, at which time endosperm formation begins. At the time of fusion the embryo sacs are long and slender (fig. 10), but soon there is an increase in the diameter of the upper end of the cavity accompanied by an apparent corresponding decrease in the length (fig. 11). This behavior allows the cytoplasm in the long endosperm cell to become shorter and more massed around the egg, and possibly accounts for the close proximity of the endosperm nucleus to the egg cell (fig. 11).

The great uniformity of the position of the endosperm nucleus with reference to the egg, the large refractive nucleolus which it contains, and the dense, apparently homogeneous condition of the chromatin of this structure cannot fail to attract the attention of the observer. Also, the nucleus and the nucleolus of the egg cell are very constant in size and appearance.

In addition to the egg apparatus, there are, in some of the embryo sacs, many densely stained bodies (possibly leucoplasts) that have the appearance of food particles. They seem to collect near the micropylar end and are so numerous in some instances as to obscure the regular cells of that region.

There may be two, three, or four (generally three) antipodal cells arranged in a single longitudinal row in this species (figs. 10 and 11). They do not disorganize early, as in *E. annuus* and *E. strigosus*, but remain in the lower end of the embryo sac for some time. Each cell is separated from the adjacent one by a definite wall which is put down after the free cell formation. A four-celled row arises when a nucleus of one of the cells divides and a cell plate is put down. The latter is an unusual condition in these antipodals, for while multinucleated cells are common, they seem to come about by the direct division of the nuclei, since no mitotic figures have been observed. The tendency is then to have three cells, one or all of which may be multinucleated. In figures 10 and 11 it will be noticed that the cells

of the integument bordering on the antipodal region are not part of the nutritive layer that is present around the upper part of the sac; instead, they are cells like those of the integument in general. This tissue disorganizes in the presence of the row of antipodal cells which may use some of the food for themselves but also absorb and transport some of it to the developing structures in the upper end of the sac.

In such an embryo sac as has just been described and as shown in figure 11, there is evidence that fertilization takes place in this species. The pollen tubes are very small, but there is usually a trace of their growth through the micropyle. The stain brings out characteristic disorganized tissue in this region at this age of the ovule but not earlier. While the condition described above is common to many ovules, the presence of the pollen tube and sperm nuclei in the embryo sac is hard to find. Figure 11 shows such a case, in which one nucleus is still in the pollen tube; and in the adjacent section there is a rather dark object which was taken to be the other sperm nucleus applied to the side of the nuclear membrane of the egg (fig. 11). Land (1900) in his account of double fertilization in this species shows the nucleus from the pollen tube, which is in contact with the egg nucleus, almost as large as the egg nucleus itself and the one in contact with the endosperm nucleus larger yet. The present investigation has not revealed anything to substantiate Land's account. The sperm nuclei are very small, as shown in figure 11. Also, in the well authenticated cases of double fertilization, the second sperm nucleus unites with the two polar nuclei at the time that they are fusing, and it seems that, after this first fusion, at which time the nuclear membrane becomes thicker, a second fusion would be impossible. Therefore, as regards the double fertilization, could it not be that those nuclei are the two polar nuclei just late in fusing? Judging from the drawing that was made in Land's investigation and the material studied by the present writer, such a condition seems plausible.

Endosperm and embryo formation begin immediately after fertilization. The building up of the endosperm is quite rapid, and soon there is a large amount of cellular food tissue around the developing embryo. As the latter grows, however, all the endosperm as well as the nutritive layer and adjacent tissue of the integument is consumed, and when the achene is mature all of the food is stored within the cotyledons of the embryo.

SPECIAL REFERENCE TO THE DEVELOPMENT OF THE ANTIPODAL CELLS OF ERIGERON PHILADELPHICUS

A discussion of the embryo sac of *E. Philadelphicus* would not be complete without special reference to the unusual development of many of the antipodal cells. The uniformity which characterizes the egg apparatus and endosperm nucleus is in striking contrast to the lack of regularity which attracts the attention of the observer when the lower end of the sac is studied. Figures 10 and 11 show embryo sacs with the antipodal cells as might be expected, but the following presentation will show what variation from the orthodox type is to be found.

It is the lower antipodal cell that shows most in this peculiar behavior. In cases where it departs from the type shown in figures 10 and 11, this cell may become so much enlarged that the entire embryo sac presents the appearance of two embryo sacs with a common central antipodal region. The other antipodal cells remain the usual size and therefore form a constricted area between the two enlarged ends. Such development is a decided departure from the type of antipodal cells found in the two former species, where early disorganization of these cells is the rule.

A few figures representing a series that may be found in the material that has been studied will be described to show some of the peculiar conditions that exist. In order to conserve space the upper end of the embryo sac has been omitted from the drawing where possible.

Figure 12 shows the antipodal region of an embryo sac at the time of fusion of the polar nuclei. A well formed egg cell and the two synergids are present in the micropylar end, all of which is normal for the species. As has been said, binucleated cells are common in the antipodals, but it is the enlargement and the particles that are noticeable here. The food granules around the egg apparatus have already been mentioned and the similarity of the two cannot be overemphasized. The protoplasm is shrunken somewhat, but in most cases these cells have large vacuoles and cytoplasm evenly distributed around the wall.

Figure 13 is from an embryo sac at the time of fertilization. The lower cell of the antipodals in this case was larger than the cavity in which the egg and endosperm nuclei were found. The

cell is unique for its content of fourteen large, well defined nuclei, all rather closely massed. The failure to find any mitotic figure in any of the numerous cases of unusual development leads the writer to think that these nuclei arise by direct division.

A little later stage is shown in figure 14, a time when a young embryo is developing in the upper end of this embryo sac. There is some shrinkage in the upper end of the antipodal region, which may mean that those cells in the constricted area are being absorbed into the embryo sac. The integument tissue bordering the structure shows considerable deterioration. There is as much disorganization of the surrounding tissue in the region of the constriction of the antipodal cells as there is about the lower enlarged region. Yet these cells never become large like the basal one. Chamberlain (1895) found that the basal antipodal cell of the embryo sac of *Aster novae-angliae* made a space for its growth by breaking through into the integument tissue. Altogether it appears that the enlarging of the basal cell cannot be due to space made available when the integument tissue is consumed, or all the antipodal cells would enlarge. It seems natural for the lower cell to grow at the expense of the surrounding tissue, and any tendency of a cell of this nature to persist and develop is important since it offers an opportunity to study the antipodal region in a different light than is possible in cases where these cells disorganize early. Numerous theories about these cells have been advanced which need verification or rejection, and it is possible that such cases as the one under consideration may prove valuable. The nuclei of the enlarged cell (fig. 14) are characteristic, especially the upper three. The one at the top resembles the egg nucleus in this species, and the next two are very much like polar nuclei. The similarity is very noticeable where both can be observed on the same slide.

Figure 15 shows the abortion of the upper end of the embryo sac and an extra layer of nutritive tissue occuping the space that is ordinarily taken up by the egg apparatus and the endosperm cell. There is only a line of disorganized material where the embryo should develop. The lower end is filled with four cells, and, judging from the position they have with reference to the nutritive or tapetal-like layer of the integument, they are the regular antipodal cells. Of the two nuclei of the lower cell, one resembles the endosperm nucleus and the other an egg

nucleus. The latter is contained in a somewhat pear-shaped area of protoplasm and suggests an egg cell.

The persistence of the enlarged basal cell is evident in figure 16. The embryo has grown until the cotyledons are about half as long as the hypocotyl. Only a small amount of endosperm is left. Just a trace of an antipodal cell in the constricted area is to be seen. About one-third of the integument tissue around the entire embryo sac has been consumed. Yet the lower antipodal cell persists and contains five distinct nuclei and some cytoplasm.

In conclusion it might be said that the same end comes to all of these cells regardless of how much they may have developed beyond that which is ordinarily seen in this region. All are used in the building of the embryo, which consumes everything in the way of endosperm, antipodal tissue and integument, until finally the full-sized embryo, covered by the dry, crushed cell walls of the ovule, fills all the space within the achene.

Chamberlain (1895) found that the antipodal region of the embryo sac of *Aster novae-angliae* exhibited a varied and much developed area of cells. He felt positive that one particular nucleus was an egg nucleus surrounded by a mass of protoplasm just as in the egg cell.

Palm (1914) reached the conclusion that the extensive development in the antipodal region of *Aster novae-angliae* and *Solidago serotina* was due to the persistence and growth of the lower megaspore of the tetrad.

Opperman (1904) reasoned that an egg nucleus and an endosperm nucleus which she found toward the antipodal end of the embryo sac were derived from one of the three antipodal cells. Fertilization of this egg cell is figured and the conclusion drawn that the antipodals are potential gametes.

Strasburger thought that the antipodal cells were homologous with the gametophyte of the gymnosperms. Except as to origin, in order for this homology to be complete the antipodal region would have to produce a nucleus that when fertilized would develop an embryo just as the gymnosperm gametophyte bears archegonia, an egg cell of which when fertilized produces an embryo.

As has been shown, the extra development in *E. Philadelphicus* is from the lower antipodal cell. Only the lower mega-

spore survives and functions in the embryo sac development; so the enlarged area could not be due to a persistent megaspore, as Palm (1914) concluded from his study on *A. novae-angliae* and *S. serotina*.

The writer feels that the extensive development of an antipodal cell is worthy of careful consideration. We are dealing with gametophyte tissue, with nuclei that arise just as the egg nucleus does, and it may be that one of them or a derivative of any one is a potential gamete. We can never be sure, however, until an actual case is found in which an embryo is obtained from a fertilized nucleus of this region. Until there is more evidence, it is best to look upon these antipodals as cells that have great persistence and power of growth but eventually have their activity ended by being incorporated into the more important structure—the embryo.

ANOMALY IN THE DEVELOPMENT OF THE EMBRYO SAC OF ERIGERON PHILADELPHICUS

An unusual embryo sac is shown in figure 17. A single longitudinal row of seven cells appears instead of the ordinary embryo sac. This drawing was made from the ovule adjacent to the one for figure 11, from which the fertilization stage was taken. Each of the two cells near the micropylar end has one poorly defined nucleus. In addition to the nucleus in the upper cell, there are a few of the densely stained bodies that have been mentioned before as food particles. The third cell is larger than the others and has a large nucleus which does not seem very well defined. No distinct nucleolus appears in it. The content is scattered throughout the structure. This third cell is larger and better supplied with cytoplasm than any of the others, although it has two large vacuoles, such as are common to all members of the row. The other four cells arranged in the row toward the chalazal end show a tendency toward the multinucleated condition. All have at least two, more or less clearly defined nuclei, and the two that are found in the middle cell of the row are most outstanding of all. In the antipodal region the nuclei are small and indistinct.

A pollen tube is present in the upper end of this embryo sac. The characteristic growth through the micropyle can be traced down into the enlarged area where it shows as a small, slender

tube along the side of the cell (fig. 17). The content is not distinct; yet a few very small bodies can be seen. The direction of growth indicates that it is passing the first cell on its way toward the chalazal end. It seems that the pollen tube is responding to a stimulus from a cell farther down in the row of cells, possibly the third one, which might be assumed to correspond to the ordinary egg cell, leaving the two upper ones to take the place of synergids. Such an assumption would explain the tendency of the pollen tube to grow on past the nucleus of this first cell. An entire embryo sac can be constructed if we grant that the two nuclei in the middle cell are polar nuclei and the three lower cells the antipodals.

FAILURE OF YOUNG DECAPITATED HEADS OF ERIGERON PHILADELPHICUS TO PRODUCE SEED

Some of the heads of flowers of *E. Philadelphicus* were decapitated in the manner described for *E. annuus* and *E. strigosus*. Specimens of several ages were selected, some before the flowers had opened and a few after the corolla tubes had been expanded for about two days. The latter were watched until the head showed signs of seed dispersal and were then gathered along with ripe seed from inflorescences that had grown normally. Figure 18 represents an achene with the pappus attached that is a fair representative of either the normal achene or one from a decapitated open head. Fertilization had already been accomplished in these open flowers; so, except for a wound on a part of the flower that was destined to fall away soon, there was no abnormal condition introduced.

With such a specimen as is represented in figure 19, however, conditions have been different. This drawing was made from an achene taken from a head that was decapitated before the flowers had opened and consequently before fertilization had taken place. A drawing of any of the various ages of heads that were used in this part of the experiment would show the same kind of an undeveloped achene. The tissue that is to become the seed coat grows to nearly normal size, but there is nothing inside to fill out the cavity. Evidently fertilization and its consequent development were prevented. Observations from time to time, when these heads should be ripening on the plant, show a development far below normal. The ovaries do not increase in

size to make a large well rounded head. Also, there is a withering of the entire head, and no indication that the achenes are going to be dispersed in the normal way. After a sufficient length of time has elapsed for normal achenes to ripen, these decapitated heads can be gathered and examined. However, no vigilance is necessary in order to obtain them, because they die away gradually and are never dispersed as are normal seeds.

It is clear that this experiment strengthens the result obtained with the preceding species. When young heads are decapitated in either *E. annuus* or *E. strigosus*, the condition is the same as with *E. Philadelphicus* where the well opened heads are used or where the seeds are allowed to develop normally on any one of the plants. No drawing was made for the achenes obtained when the apogamous species were used, and it will suffice to say that they compare favorably with the achene shown in figure 18. The clear cut variation in behavior with the different experimental plants coupled with the morphological differences already explained make a consistent demand for recognition of apogamy in *E. annuus* and *E. strigosus*, but of normal development in *E. Philadelphicus*.

SUMMARY

Erigeron annuus. There is no tetrad of spores formed from the megaspore mother cell of *E. annuus*. Fertilization was not observed in this plant and is not necessary in the setting of seed. Decapitated heads of various ages develop achenes with viable embryos just as normal heads.

Erigeron strigosus. *E. strigosus* duplicates *E. annuus* in embryo sac development and experimental behavior.

Erigeron Philadelphicus. A tetrad of spores is formed from the megaspore mother cell of *E. Philadelphicus*. Also, fertilization is necessary for the further development of the ovule. Decapitated heads of stages before the flowers opened did not continue to develop but withered gradually. Unusual development may be observed in the antipodal region of the embryo sac. An anomaly in which the embryo sac is represented by a longitudinal row of seven cells is described.

The writer is grateful to Dr. David Myers Mottier, of the University of Indiana, for his appreciative interest and helpful criticism during the progress of this work.

INDIANA UNIVERSITY,
BLOOMINGTON, INDIANA.

Literature cited

Beer, R. 1912. Studies in spore development. 2. On the structure and the division of the nuclei in the Compositae. Ann. Bot. **26:** 705–726. Pl. 66, 67.

Chamberlain, C. J. 1895. The embryo sac of *Aster novae-angliae*. Bot. Gaz. **20:** 205–212. Pl. 15, 16.

Holmgren, I. 1916. Apogamy in the Gattung *Eupatorium*. Svensk. Bot. Tids. **10.**

Juel, H. O. 1898. Parthenogenesis bei *Antennaria alpina* (L.) R. Br. Bot. Centralbl. **74:** 369–372.

———. 1900. Vergleichende Untersuchungen über typische und parthenogenetische Fortpflanzung bei der Gattung *Antennaria*. Kungl. Svensk. Vet. Akad. Handl. **33:** 1–59. Pl. 1–6, 5 fig.

———. 1904. Die Tetradenteilung in der Samenlage von *Taraxacum*. Ark. Bot. Sven. Vetensk. Akad. **2:** 1–9.

———. 1905. Die Tetradenteilung bei *Taraxacum* und andern *Chicoraceen*. Kgl. Sv. Vet. Akad. Handl. **39:** 209–226. Pl. 1–3.

Land, W. J. G. 1900. Double fertilization in the Compositae. Bot. Gaz. **30:** 252–260. Pl. 15, 16.

Murbeck, S. 1904. Parthenogenesis bei den Gattungen *Taraxacum* und *Hieracium*. Bot. Not. **1904:** 285–296.

Opperman, Marie. 1904. A contribution to the life history of *Aster*. Bot. Gaz. **37:** 353–362. Pl. 14, 15.

Osawa, I. 1913. Studies on the cytology of some species of *Taraxacum* Archiv. Zellforsch. **10:** 450–469. Pl. 37, 38.

Ostenfeld, C. H. 1904a. Zur Kenntniss der Apogamie in der Gattung *Hieracium*. Ber. Deut. bot. Gesellsch. **22:** 376–381.

———. 1904b. Weitere Beiträge zur Kenntniss der Fruchtenwicklung bei der Gattung *Hieracium*. Ber. Deut. bot. Gesselsch. **22:** 537–541.

———. 1906. Experimental and cytological studies in the genus *Hieracium*. 1. Castration and hybridization experiments with some species of *Hieracium*. Bot. Tidsskrift. **27:** 225–248. Pl. 1.

Palm, B. 1914a. Zur Embryologie der Gattungen *Aster* und *Solidago*. Acta Horti Bergiani **5:** 1–18. 27 fig.

———. 1914b. Ueber die Embryosackentwicklung eineger Kompositen. Svensk. Bot. Tids. **8.**

Raunkiaer, C. 1903. Kimmdannelse uden Befrugtning hos Molkebötte (*Taraxacum*). Bot. Tidsskr. **25:** 109–140. Rev. Bot. Centralbl. **93:** 81–83.

Rosenberg, O. 1906. Ueber die Embryobildung in der Gattung *Hieracium*. Ber. Deut. bot. Gesellsch. **24:** 157–161. Pl. 11.

————. 1907. Experimental and cytological studies in the genus *Hieracium*. 2. Cytological studies on the apogamy in *Hieracium* Bot. Tidsskrift **28**: 143–170. Pl. 172, 8 fig.

————. 1909. Zur Kenntnis von den Tetradenteilungen der *Compositen*. Svensk. Bot. Tids. **3**.

————. 1912. Ueber die Apogamie bei *Chondrilla juncea*. Svensk. Bot. Tids. **6**.

Sears, P. B. 1917. Amiotic parthenogenesis in *Taraxacum vulgare* (Lam.) Schrk. and *Taraxacum laevigatum* (Willd.). D.C. Ohio Jour. Sci. **17**: 97–100.

Stork, Harvey E. 1920. Studies in the genus *Taraxacum*. Bull. Torrey Bot. Club **47**: 199–210. Pl. 6, 7.

Winkler, H. 1908. Ueber Parthenogenesis und Apogamie in Pflanzenreich. Prog. Rei Bot. **2**: 293–454. Fig. 1–14.

Explanation of plates

PLATE 32

Fig. 1. Section of young ovule of *E. annuus* showing nucellus, *n;* megaspore mother cell, *Meg. M.C.;* and beginning of the integument, *int.*

Fig. 2. Section of nucellus of an ovule of *E. annuus*. The integument, *int*, has grown up around the nucellus and forms the micropyle, *mic*. The large cell is the morphological megaspore, *meg*.

Fig. 3. Section of nucellus, *N*, of *E. annuus* showing the disorganized nucellar tissue. The morphological megaspore, *meg*., has not yet divided to form an embryo sac with two nuclei.

Fig. 4. Embryo sac, *emb*., of *E. annuus* with two nuclei. Nucellar tissue, *N*, disorganized at the upper end.

Fig. 5. Embryo sac, *emb*., of *E. annuus*, with four nuclei. Unorganized nucellar tissue, *N*, is shown around the lower end of embryo sac.

Fig. 6. Embryo sac of *E. annuus* at the time of fusion of the polar nuclei, *pol*. Micropyle, *m;* morphological egg, *e;* synergids, *s;* antipodals, *ant.*

Fig. 7. Young embryo of *E. annuus* showing suspensor, *s*, and two cells. The lower cell is binucleate. The nuclei tend to be binucleolate.

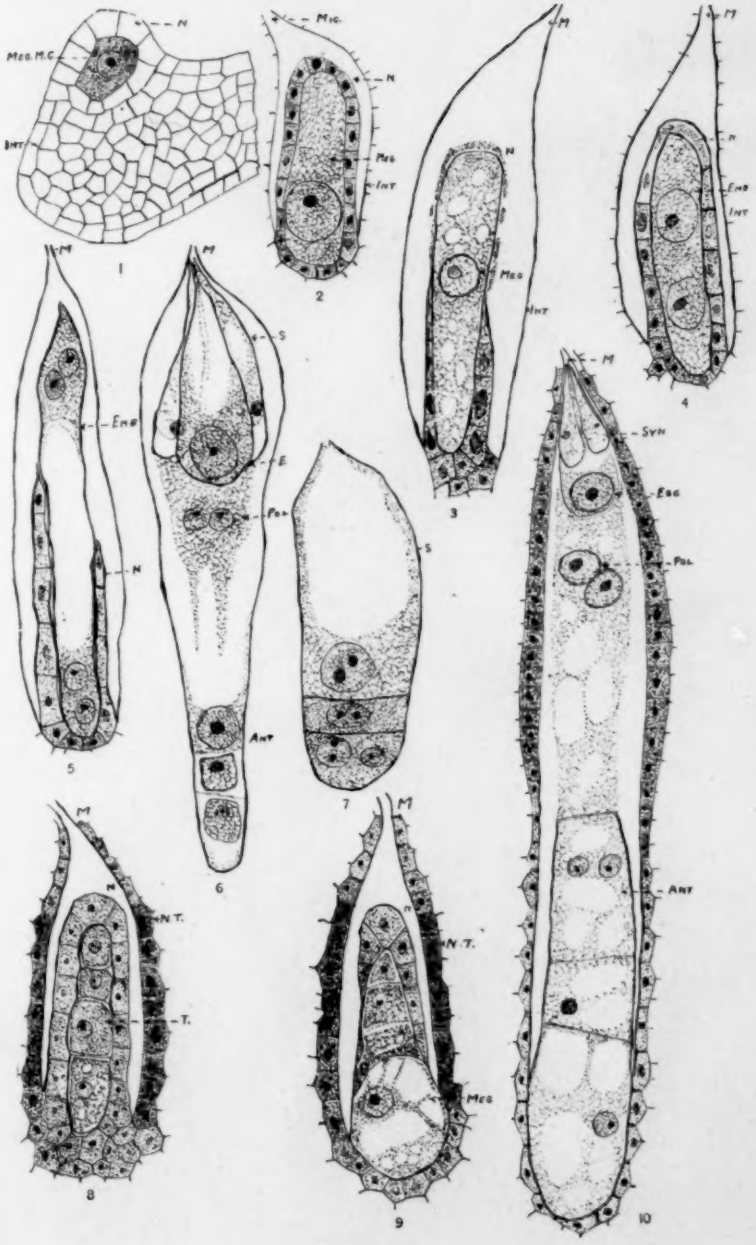
Fig. 8. Section of the nucellus, *n*, and nutritive layer of tissue, *n.t.*, of an ovule of *E. Philadelphicus*. A tetrad of megaspores fills the nucellus.

Fig. 9. Section of the nucellus, *n*, and nutritive layer, *n.t.*, of *E. Philadelphicus* showing the lower megaspore consuming the other three and also the nucellus.

Fig. 10. Embryo sac of *E. Philadelphicus* at the time of fusion of the polar nuclei, *pol*. Three antipodals, *ant*., and the egg apparatus, *egg, syn*., are present. The tapetal-like layer of the integument is distinct down to the antipodal region.

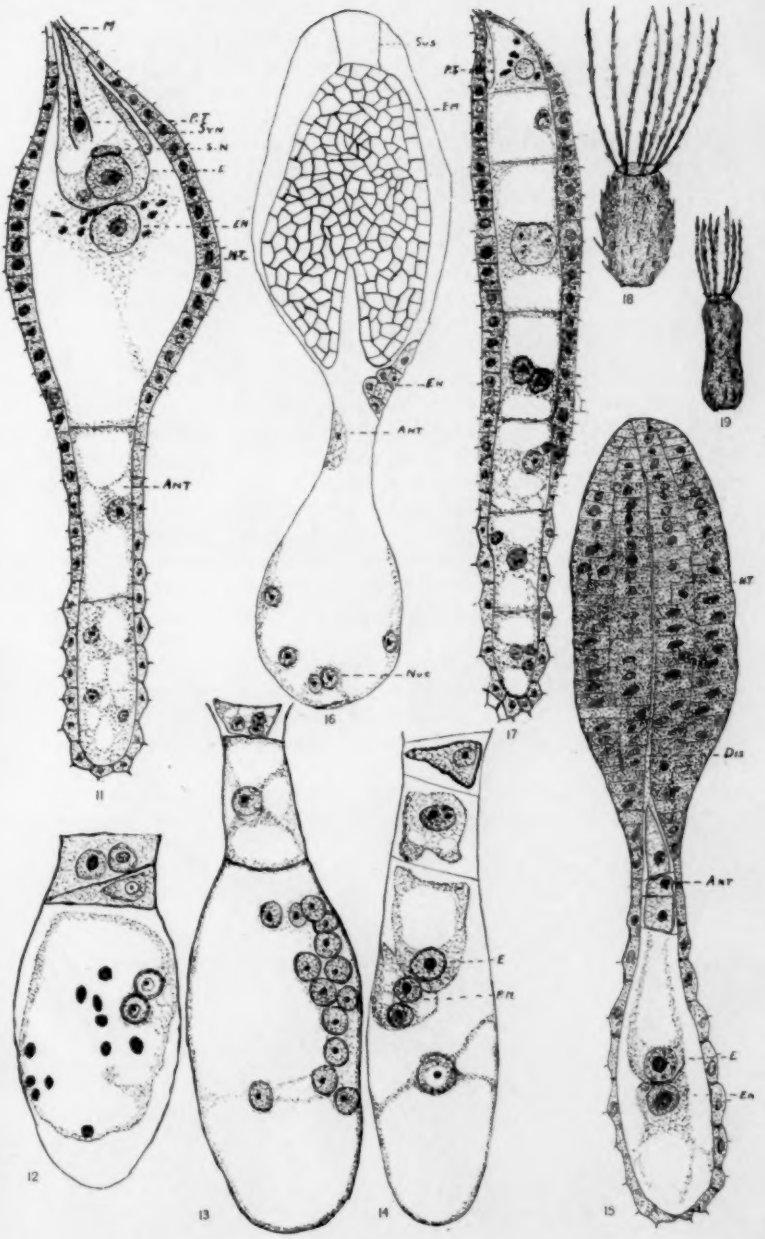
PLATE 33

Fig. 11. Embryo sac of *E. Philadelphicus* at the time of fertilization. Pollen tube, *p.t.*, containing one nucleus; egg, *e*, with sperm nucleus, *s.n*., applied to the nuclear membrane; synergid, *syn.;* endosperm nucleus, *en*., with food bodies around it; *m*, micropyle; and *ant*., antipodal region.


N
Meg. M.C.
INT
Mic.
N
Meg
INT
M
N
Meg
INT
M
N
END
INT
M
END
N
1
2
3
4
M
S
E
Pol
ANT
5
6
7
M
SYN
Egg
Pol
ANT
M
N
N.T.
T.
M
N
N.T.
Meg
8
9
10

ERIGERON

UNIV. OF MICH.


M
P.T.
SYN
S.N
E
EN
NT.
ANT
11
SUS
PS
EM
EN
ANT
NUC
16
17
18
19
aT
DIS
ANT
E
EN
15
12
13
E
EN
14

ERIGERON

UNIV. OF MICH.

Fig. 12. Antipodal end of embryo sac of *E. Philadelphicus*, at the time of fusion of the polar nuclei, showing the lower, much enlarged cell containing two nuclei and several food particles. The two cells above it are the antipodal cells in the constricted area of the embryo sac.

Fig. 13. Antipodal end of embryo sac of *E. Philadelphicus* at the time of fertilization showing the much enlarged lower cell containing fourteen free nuclei. The cell nearest the egg apparatus end of the embryo sac is being absorbed.

Fig. 14. Antipodal cells of *E. Philadelphicus* at a time when a young embryo is present in the upper end of the embryo sac. The cells in the constricted area are being absorbed. The enlarged basal cell has four nuclei, the upper one of which resembles an egg cell, *e*, and the two just below it resemble polar nuclei, *p.n.*

Fig. 15. Section of an embryo sac of *E. Philadelphicus* showing a characteristic antipodal region of four cells. Of the two nuclei of the basal cell, the upper one, *e*, resembling an egg nucleus and the one just below it an endosperm nucleus, *e.n.* The egg apparatus is aborted and shows only as a line of disorganized material, *dis.* An extra layer of tapetal-like tissue, *n.t.*, has developed where the egg apparatus should be.

Fig. 16. Section of an embryo sac of *E. Philadelphicus* showing the enlarged antipodal cell still present after an embryo, *em.*, has grown large enough to fill the egg apparatus end of the sac. A portion of endosperm, *en.*, is still present and also a remnant of an antipodal cell, *ant.*, of the constricted area between the two ends.

Fig. 17. Anomaly in the development of the embryo sac of *E. Philadelphicus*. A single longitudinal row of seven cells composes this embryo sac. A pollen tube, *p.t.*, is present in the upper end.

Fig. 18. Normal achene developed in *E. Philadelphicus* when an open head is decapitated.

Fig. 19. Empty undeveloped achene of *E. Philadelphicus* which results from unopened decapitated heads. Evidently fertilization is prevented.

The production of intumescences in Transparent apple by ethylene gas as affected by external and internal conditions

RAYMOND HAROLD WALLACE

(WITH TWO TEXT FIGURES AND PLATE 34)

Dating from the classic work of Nägeli in 1893 on oligodynamic phenomena a vast mass of physiological data has been accumulated upon the effects of extremely small amounts of various organic and inorganic substances in modifying the growth and inducing specific responses of various types in plants. Sensitiveness to certain compounds in a degree not before imagined has been demonstrated. It has been shown recently that mere traces of boron, zinc, manganese, and other elements may be essential for the growth and maturity of some species. We now know that plants respond to concentrations of toxic and stimulating substances which are far below the range of measurement by any *in vitro* physical or chemical means at our command. Neljubow (1901), Crocker and Knight (1908), and others have shown the sensitiveness and the extreme variation in the reactions of plants to minute amounts of ethylene, and I have given a preliminary report on the production of intumescences on woody twigs by low concentrations of this gas.

LITERATURE REVIEW

Physiological effects of ethylene

That ethylene is a substance which can induce a wide variety of effects in living organisms is well known. The earlier data relate primarily to the toxic effects shown by etiolated seedlings, roots, and other plant parts when exposed to very low concentrations of this gas. Neljubow (1901) reported that the epicotyls of etiolated seedlings of pea, *Vicia sativa*, *Ervum lens*, *Lathyrus odoratus*, and *Tropaeolum* grow horizontally in the presence of ethylene, acetylene, or illuminating gas. A concentration of 1 part of ethylene to 1,000,000 parts of air affected the direction of growth, while 1 part of ethylene to 40,000 parts of air caused the death of the seedlings. Crocker and Knight (1908) found

the buds and flowers of carnations to be very susceptible to injury by ethylene gas. They report that the closing of the flowers of the carnation occurred in a concentration of 1 part of ethylene to 2,000,000 parts of air. A concentration of 1 part of ethylene to 40,000 parts of air was lethal to young buds and prevented the opening of buds already showing petals. E. M. Harvey (1913), working in the same laboratory, found that the leaves of *Ricinus communis* exhibit what he calls nastic droopings in a concentration of 1 part of ethylene to 10,000,000 parts of air. These results show that some plants or plant parts may be seriously injured or killed by extremely low concentrations of ethylene, while others may exhibit definite nastic or tropic responses.

Ethylene gas induces hypertrophy and hyperplasia in the tissues of some plants. Harvey and Rose (1915) found that swellings developed in the root cortex of seedlings of *Catalpa speciosa* and *Ailanthus* after exposures of from five to twenty-one days to various concentrations of ethylene or illuminating gas. Similar tests with *Gleditschia* gave no swellings. Both hypertrophy of the cortical parenchyma and hyperplasia of the phellogen were said to be involved. Abnormal tissues also developed in the cortex of *Hibiscus*, *Syringa*, *Diervilla*, *Ricinus*, *Ulmus*, croton, and pear, when only a small current (about 40 cc. daily) of illuminating gas was used. They thought that ethylene was mainly responsible for these formations.

Doubt (1917) extended the work by using two- or three-year-old trees and allowing various concentrations of the gas to flow around the roots continuously for from fifteen to ninety days. Her results were similar to those of Harvey and Rose. She reports all proliferated tissue to be external to the vascular cylinder.

More recently ethylene has been used to hasten bleaching or the disappearance of chlorophyll from various plant parts. Denny (1924) found that the yellowing of lemons was greatly hastened by exposing them to traces of ethylene. He also found that ethylene produced a definitely increased rate of respiration when in concentrations as low as 1 part to 1,000,000 parts of air. R. B. Harvey (1925) found that ethylene hastened the disappearance of chlorophyll from the leaves of celery. Rose (1925) found a similar effect in the case of green fruits of

tomato. He also reports using ethylene successfully to induce more rapid sprouting of dormant potato tubers.

Luckhardt and Carter (1923) established the fact that ethylene gas, when inhaled by animals, including man, has strong anaesthetic properties. A mixture of 90 per cent of ethylene and 10 per cent of oxygen anaesthetized all animals studied, but the time required for complete anaesthesia was found to vary somewhat with the individual and species. The characteristics of ethylene anaesthesia are somewhat similar to those produced by nitrous oxide, but are very different from those produced by ether or chloroform. Ethylene seems to cause no marked stimulation or depression of the vagus centers. "Respiration" and blood pressure remain almost normal. It is now used quite extensively in some medical institutions.

One of the most peculiar of the physiological effects reported for ethylene gas is that noted by Fujii (1925). He observed, while making a study of *Isoetes*, that occasionally the antheridia liberated their sperms with great readiness. At other times it was impossible for him to induce their escape by any means at his command. Numerous experiments finally convinced him that the slight amount of illuminating gas set free in the room at the time he lighted the gas lamp for his microscope was responsible for the effect. Further experiments indicated that in this case also ethylene is the active constituent of the illuminating gas.

The formation of intumescences in plants

Pathological outgrowths of the type known as intumescences are found to occur in numerous kinds of plants and plant tissues. While these malformations most frequently appear on leaves and stems, they are not uncommonly found on roots and may even occur on flowers and fruits. Numerous hypotheses have been advanced as to the conditions which induce or favor the production of these peculiar outgrowths. Sorauer, in one of his early papers (1886) concludes, as a result of extensive experiments on intumescences in leaves, that their formation is to be regarded as a symptom of a metabolic disturbance due to an excess of water during a period of low assimilation. Later (1899) he repeats this hypothesis, but stresses also the contributory effect of a high temperature. He says that a high temperature plus an excessive water supply in the case of plants

whose "assimilative activity" is low because of weak illumination, will result in the production of intumescences. Quoting him directly: "Since in two cases (*Ficus elastica* and *Impatiens Sultani*) I have already succeeded in producing intumescences by such means, the appearance of such structures may now be regarded with certainty as a symptom of an abnormal rise in temperature and excessive water supply."

Excessive water supply in the soil or in the atmosphere is given by many investigators as the determining condition in intumescence formation. Atkinson (1893a) reports a very interesting oedema upon tomato plants which were grown in a forcing house. These plants exhibited numerous elevated areas of granular appearance on the surfaces of the leaves, petioles, and stems. The appearance was so suggestive of a bacterial or fungous infection that many experiments were made before it was established that the disease was of non-parasitic origin. Atkinson concluded that the chief physiological cause for the disease was an excessive water content, due to high root pressure and low transpiration. The weak illumination and high humidity present in the experimental greenhouse permitted only a very low rate of transpiration as well as allowing only a minimal rate of photosynthesis. He was able to produce similar intumescences at will by injecting the shoots with an excess of water. He reports (1893b) a somewhat similar case of oedema on the stems and branches of apple. The elevations arose from the phellogen and were from one-eighth to one-fourth inch in length and almost as wide, and appeared as blisters on the bark of the trees. This diseased condition appeared in trees whose transpiring powers had been made low by severe top pruning, but whose root systems had not been molested and were therefore capable of rapid absorption. He concluded, in this case, that the outgrowths were due to excessive root pressure which was developed as the result of a high rate of absorption by the roots and a low rate of transpiration from the aerial parts of the plants.

Steiner (1905) found intumescences on the leaves of *Ruellia formosa* and *Aphelandra Porteana*, and concluded from experiments that they were developed in response to excessive humidity. They did not appear when the leaves were immersed in water or kept in darkness. Douglas (1907) produced a great

number of intumescences upon the stems and leaves of potato plants by enclosing the plants in moist atmospheres under bell-jars. They formed readily in either weak or strong light, but did not appear when the plants were kept in total darkness.

Intumescences in woody stems have been induced by covering them with a relatively impervious layer of paraffin oil, vaseline, etc. Wisniewski (1910) painted twigs of *Ficus australis* and *Ficus elastica* with paraffin oil and kept them in laboratory air. In a week they developed in their lenticels intumescences as much as 4 mm. in diameter. Twigs, untreated by paraffin oil, but placed under bell-jars and partially immersed in water, produced some small intumescences in their lenticels on the immersed portions of the stems, while the portions in the humid air above were free from outgrowths. From this he concluded that, while a reduction of transpiration is to be regarded as one of the main conditions influential in causing intumescences, other conditions, yet poorly understood, play a part as well.

Schilling (1915) found that the response reported by Wisniewski for two species of *Ficus* was characteristic of many species of plants. He reported that lenticel intumescences or excrescences appeared upon stems of *Aesculus hippocastanum*, *Spiraea concinna*, *Philodendron pinnatifidum*, *Ascocarpus incisa*, *Sambucus nigra*, and numerous other woody plants when the stems were painted with vaseline or paraffin oil. In general, plants having lenticels were more prone to form intumescences than those lacking them. Schilling concludes that these abnormal developments are not due to any chemical stimulus from the vaseline or paraffin oil. He regards retardation of transpiration as the chief condition involved, but also mentions the possibility that oxygen deficit may play a rôle as well. Tests indicated that the osmotic pressure of the abnormal cells was greater than that of normal cells.

Hahn, Hartley, and Rhoades (1920) reported tissue formation very similar to that noted above on woody stems as occurring on the roots of a great variety of coniferous seedlings when grown under conditions of excessive soil moisture. The disease was present in about equal degree in both strong and weak seedlings, which, they thought, indicated that it was a physiological derangement not dependent on the presence of a parasitic organism. Top pruning decreased the intensity of

the disorder while root pruning increased it. The writers were doubtful as to the major determining condition, but were inclined to think it was either a deficit of oxygen or an excess of carbon dioxide rather than an excessive sap pressure.

Special light relations, as well as an excessive supply of moisture, have been considered as favoring markedly the production of abnormal outgrowths upon plants. Although the reports concerning the influence of light are conflicting, a survey of the literature indicates that light may either favor or inhibit the response, according to the plant materials and the other experimental conditions involved.

One group of investigators has been inclined to look upon weak illumination as favorable to intumescence formation. Sorauer (1886) held that low "assimilative activity" resulting from weak illumination tends to favor the production of such malformations. He also states (1899) that intumescences form most often during the fall and winter when the assimilative activities of the plants are lowest. Likewise, Atkinson (1893a) regarded weak light as favoring intumescence formation in the stems and leaves of the tomato. He associates the production of intumescences both with the low photosynthetic activity that results from poor illumination and with the development of weak, watery tissue that occurs in low light intensity. The formation of the intumescences described by Prillieux (1892) on the leaves of the pink and by Norack (1901) on the leaves of the grape was observed to occur only in weak illumination.

Küster (1903b) produced artificial intumescences on the leaves of *Populus tremula* by floating the leaves on water. These outgrowths formed both in light and in darkness, but in strong light they appeared usually on the side of the leaf nearest the water, regardless of whether this was morphologically the dorsal or the ventral side. Their formation on the under side was considered to be associated with retardation in the rate of transpiration on that side.

Another group of investigators has found that rather strong light is essential to the production of intumescences. Dale (1901) reported that the peculiar outgrowths which often occur on the leaves, petioles, and green stems of *Hibiscus vitifolius* failed to appear in total darkness or in weak light. The quality of the light was also found to be significant, since the intumes-

cences formed under white, red, or yellow light, but failed to form under blue, green, or weak light. She pointed out that a checking of the rate of transpiration was one of the necessary conditions determining the development of intumescences, but that this condition alone was not sufficient to lead to their formation. An active rate of photosynthesis was also considered to be necessary.

Viala and Pacottet (1904) drew a similar conclusion with reference to intumescences that appeared on the leaves of grapes grown under glass, since they found by experiment that these outgrowths occur only in the presence of intense light and a humid atmosphere. Only leaves directly under the glass during periods of strong illumination were affected, while those in weak light or in the shade remained normal. Von Schrenk (1905) makes a similar statement concerning the formation of intumescences on the leaves of grape. He reported, however, that the production of intumescences on cauliflower in response to chemical stimulation by copper was independent of light intensity, and occurred throughout the range of temperature and moisture conditions existing in his experiments.

Douglas (1907) held the view that light favors the production of intumescences on potato plants only in so far as it provides for the normal processes of metabolism and growth of the stems and leaves. The failure of intumescences to appear in darkness was considered to be due to a reduction of the effective water content, owing to an increased growth in length.

The data so far reviewed indicate clearly that several of the conditions comprising the physical environment may be very influential in the production of intumescences on many plants. Furthermore, there is abundant evidence that chemical conditions of the environment may have as great an effect in causing such proliferations or malformations in plant tissues. Haberlandt (1899) was able to induce the production of intumescences upon the thalli of *Conocephalus ovalus* and *C. suaveolens* by painting them with a one per cent alcoholic solution of corrosive sublimate. This treatment killed the cells of the hydathodes, or water excreting organs. Additional hydathodes were then formed by enlargement of the cells of the underlying parenchyma. Haberlandt attributed the formation of these "compensating" hydathodes to an excess pressure which he supposed developed

through the failure of the normal organs to function. The new structures functioned as normal hydathodes until they dried up and died.

Von Schrenk (1905) induced the production of intumescences on the leaves of cauliflower by spraying them with various solutions of copper salts. The occurrence of these formations seemed not to be influenced by ordinary differences in atmospheric or soil conditions. Douglas (1907) produced similar outgrowths on leaves of young potato plants by treatment with like compounds. Old potato plants failed to form these structures, but instead exhibited brown areas of dead tissue. It is interesting to note that Steiner (1905) induced the formation of intumescences on the leaves of *Aphelandra Porteana* by placing them in a saturated atmosphere, but did not succeed in producing them by treatment of the leaves with copper sulphate, ammonium chloride, formic acid, pyromalic acid, potassium hydroxide, sodium hydroxide, and alcoholic and aqueous solutions of corrosive sublimate.

Smith (1917) tested the effect of various chemicals on the leaves of cauliflower. Ammonia and Carnoy's fixing fluid (ethyl alcohol and acetic acid) gave intumescences similar to those reported by Von Schrenk for copper compounds. When leaves with open stomata were exposed to Carnoy's fixing solution, dead areas appeared quickly and intumescences never developed; but when leaves with closed stomata were used, the production of intumescences resulted.

Injections of various chemicals into the pith and bark of various woody and herbaceous stems and roots have been shown to produce very striking cases of tissue development by cells that one would have supposed had lost the capacity of further growth and differentiation. In an attempt to control the blight of the chestnut by injections of chemicals into the bark of the trees, Rumbold (1915) discovered that cells of the soft bast possessed unsuspected powers of tissue formation. Following the treatment, isolated masses of xylem, phloem, and cambial tissues developed in the bark of the injected trees. She concluded that these abnormal tissues had developed in response to chemical stimulation by the injected substances.

Taylor (1916) extended this study of tissue formation resulting from injection of chemicals, experimenting on herbaceous

as well as woody stems. Patches of xylem, phloem, and cambium were found to arise in the region of the soft bast in the bark of *Castanea*, but the evidence which he secured indicated that the development of these tissues was a response to the mechanical injury involved in injection and to the general injury resulting from the injected substances, rather than to a specific stimulative action of the different chemicals injected, as Rumbold had assumed. He regarded this formation of abnormal tissue as the usual wound response for the chestnut. The injection of distilled water, chloroform, ammonia, lithium carbonate, copper sulphate, and picric acid into the stems of *Polygonum*, *Helianthus*, *Phytolacca*, and *Ricinus* in every case resulted in the production of abnormalities. As a result of the injection of one or more of these agents, all kinds of tissues except epidermal and lignified elements were produced. Taylor concludes that any tissue in the stem not cutinized, suberized, or lignified may be induced to proliferate, and that the reaction is probably a response to the chemical or mechanical injury, depending upon the concentration and kind of injected material, and the extent of the mechanical injury involved in the injection.

Smith (1917) was able to simulate crown-gall formation in stems of *Ricinus communis* by injecting by-products of the metabolism of the organism *Bacterium tumefaciens* into the stems of the plants. Responses were usually absent in old nodes, but the younger nodes developed intumescences in the pith. In one case he produced experimentally "a stem within a stem" by injecting the internode with ammonia, while in another a second cylinder of xylem and phloem developed within the first. This secondary xylem-phloem tissue contained tracheae, wood fibers, cambium, etc., and was divided into bundles which were separated regularly by medullary rays. He concludes that the osmotic pressure of the injected substances removed, in some manner, the inhibiting factors which hold cell proliferation in check, and thus gives rise to these abnormal tissues; and further that the crown-gall organism excretes these metabolic products continuously and thus stimulates the host to form abnormal tissue.

Some instances are known where intumescences apparently develop in response to mechanical stimulation alone. Although

Küster (1903) held that gall formation on the leaves of *Populus tremula* by *Hermandia tremulae* and *H. globuli* was probably due directly or indirectly to toxic substances excreted by the gall forming organism, Rosen (1916) concluded from an ontogenetic study of the *Phylloxera* leaf gall that a chemical theory can not explain gall formation by this organism. His evidence led him to conclude that the proliferations of tissue which resulted in the formation of the gall were induced initially by the continual sucking action of the insect.

Bayliss (1907) observed abnormal swellings upon the roots of many plants in her investigations on the galvantropism of roots and suggested that they may have arisen in response to galvanic stimuli, or to products of electrolysis. Davis (1910) investigated this point further and concluded from his results that the intumescences were a response to wounding by the thermal or chemical changes induced by the electric current.

Wolf (1918) found intumescences upon the young leaves of cabbage which had suffered injury from blowing sand, and he was able to produce similar growths in the laboratory by means of a sand blast. He thought that the exposed cells underwent oxidation reactions which altered the hydrogen-ion concentration of the tissue and thus increased the imbibitional power of the cell colloids. This greater capacity for hydration of the cell colloids was considered to cause an excessive absorption of water which resulted in the distention and protrusion of the injured tissue.

The depth of penetration and the tissues involved in intumescence production in the bark of woody stems in response to external stimuli, such as illuminating gas, ethylene, paraffin, and other agents, have attracted no small amount of attention from investigators, but the data are still much confused. I have reviewed in a recent paper (1926) the published data bearing upon the type and extent of intumescences which develop in the bark of certain woody plants in response to ethylene or illuminating gas. In that survey the cambium, portions of the phloem, the cortex in general, or the phellogen only, are reported as involved in the formation of abnormal tissues. The lack of agreement among the investigators as to the precise tissues affected and the nature of the proliferations may be due to differences in the experimental material and methods used.

An examination of the data concerning the intumescences which occur in response to agents other than ethylene or illuminating gas shows a similar situation. Küster (1903), in describing the oedema or dropsy of *Ribes aureum*, gives what may be taken as an example of the most severe and deep-seated type of intumescence found in woody stems. This disease not uncommonly develops in nature under field conditions and is then a source of much trouble to the growers of currants and gooseberries, since *Ribes aureum* is used extensively as the stock in grafts. In this disease, abnormal tissue development results in the formation of deep longitudinal rifts in the bark of the affected plant; the disease may appear on tissues from one to several years old. All living elements from the cambium outwards may undergo hypertrophy, and in extremely severe cases even the cells of the young xylem may be affected. The walls enlarge at the expense of the cell contents, and the cells become long and sac-like. They are rich in water, but poor in all other materials. In some cases, when maceration is complete, the cells become detached from each other and form a mass of loose friable granules. The bast tissue which does not undergo hypertrophy is torn apart radially and pushed out by the enlarging tissues beneath. Oedemas similar to this have been described by Sorauer (1892) on *Pyrus communis* and by Molz (1906) on *Vitis*. Abnormal nutrient relations have been suggested as the cause.

The oedema on *Pyrus malus* described by Atkinson (1893b) was of relatively superficial origin. The epidermis was raised in blister-like patches by large vacuolated cells below, which had arisen by proliferation of the phellogen. The other tissues of the stems remained normal. The lenticel excrescences which Wisniewski (1910) induced in the stems of *Ficus elastica* and *Ficus australis* by painting them with paraffin oil extended below the phellogen. They arose by division and enlargement of the cells of the phellogen and somewhat deeper portions of the cortex.

Schilling (1916) extended the method of Wisniewski to a study of a great number of woody plants and found considerable variation in the response exhibited by the various species. Under his experimental conditions almost all kinds of plants developed excrescences in the lenticels, but *Ginkgo*, *Sophora*,

Ilex, and *Solanum* failed to do so. Division and enlargement occurred in almost all elements of the stems except the epidermis, cork, xylem, and bast. The sieve tubes and companion cells failed to enlarge or divide and were crushed by the pressure exerted by the other, enlarging cells. The intumescences extended to, and affected the cambium in *Ribes*, *Sambucus*, *Aesculus*, *Robinia*, and *Clerodendron*.

Definite data relative to the depth of origin of the intumescences in the lenticels of the roots of conifers are not given by Hahn, Hartley, and Rhoades (1920). The fact that death sometimes resulted when the infections were severe indicates that deep-seated tissues were involved.

MATERIALS AND METHODS

I have studied especially the production of intumescences on woody stems of *Pyrus malus* (apple, variety Transparent) and made a few general experiments on apple (variety Williams) and *Populus deltoides*. Branches from 1 to 1.5 cm. in diameter at the base were cut from trees growing out of doors. These shoots were then cut into short lengths (10–15 cm. long) and the latter were grouped into sets, each of which included a graded series of sizes from 1.5 cm. in diameter down to slender terminal twigs. I shall call such pieces cuttings. There was some variation in the number of cuttings to a set in the different experiments; but the number was never less than five nor more than twenty. The results reported in my first paper were obtained during the winter period from September to March. The work has now been extended over the entire year.

The oxygen and nitrogen used were obtained from the Linde Air Products Company and were from 98 to 99 per cent pure. The ethylene was of the quality sold by the Industrial Alcohol Company for use as an anaesthetic, and it proved to be of very high grade, from 97 to 99 per cent being absorbed by bromine water. The carbon dioxide was of the type used for preparing carbonated beverages, 99 per cent being absorbed by potassium hydroxide.

As gas chambers for the cuttings, I at first used twenty-liter bell-jars inverted in shallow pots full of sand. One and one-half liters of quartz sand were poured into each jar and water was added until the moisture content was about the same as that

commonly used for rooting cuttings. The jars were closed with ground-glass plates on their tops. The condensation of water vapor inside the jar usually maintained a water seal between the ground-glass plate and the rim of the jar. The volume of ethylene required to make the concentration desired for a test was collected in a suitable vessel, such as a glass tumbler or a battery-jar, by water displacement in a pneumatic trough. The vessel was then closed by a glass plate of suitable size, and the vessel with its cover was removed from the trough and placed in the experimental jar with the cuttings. After the ground-glass plate had been put in place over the mouth of the bell-jar, the gas was liberated by pulling a small cord which had previously been attached to the cover of the enclosed vessel. In all experiments the pieces of stems were placed with the basal end downward in contact with moist sand or filter paper as if for rooting experiments.

To obtain gas mixtures of more accurately known composition for testing the extremely low concentrations as reported, the apparatus described above was modified in several details. The ground-glass surfaces of the glass plate and the rim of the bell-jar were coated with castor oil, and a heavy weight was placed on the plate after it had been put in place. This modification reduced the chances of any possible interchange of gases between the interior of the bell-jar and the surrounding atmosphere. All quantities of ethylene or mixtures of ethylene and air which were used to make the desired concentrations were introduced into the bell-jars by means of a gas burette and mercury leveling bulb, through tubulations in the bell-jars. The tubulations were supplied with ground-glass stopcocks. It was found convenient to use ethylene-air dilutions in making the low concentrations. These were made in the following manner: Tubes supplied with ground-glass stopcocks were sealed into the mouths of twelve-liter bottles. A 120 cc. sample of ethylene was then forced into the first bottle by means of the gas burette and mercury leveling bulb, thus forming in the bottle a mixture composed of 1 per cent of ethylene and 99 per cent air, by volume. After a twenty-four hour period a 120 cc. sample of this ethylene-air mixture was withdrawn and added to a second bottle, thus forming a concentration of approxi-

mately 1 part of ethylene to 10,000 parts of air.[1] After a further equal interval of time a similar volume was drawn from this bottle and added to a third bottle, thus forming a dilution of 1 part of ethylene to 1,000,000 parts of air. These three supply bottles were then used to make the dilutions desired in the twenty-liter bell-jars in which the cuttings were exposed to the gas. For example, the addition of 200 cc. of the gas mixture from

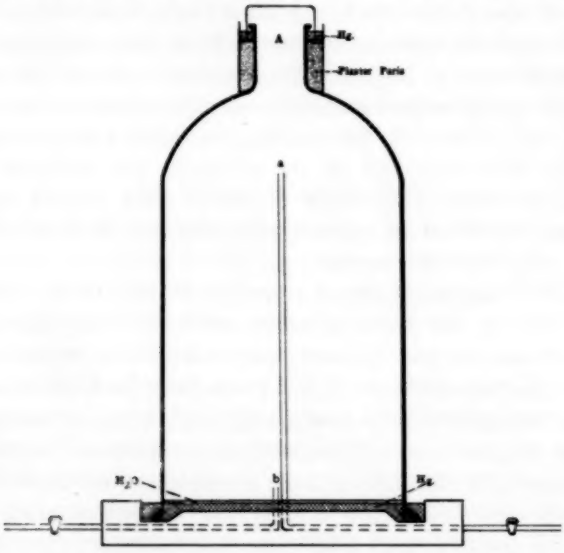


Fig. 1. Optical section of twenty-liter bell-jar and cement base used as experimental chamber for controlling the composition and rate of flow of gas mixtures supplied to the cuttings. (For full explanations see text.)

one of these bottles to a twenty-liter bell-jar formed a concentration in this jar which was one-hundredth as great as was present in the bottle from which the sample was drawn. Concentrations greater than 1 part of ethylene to 10,000 parts of air were made directly with pure ethylene by means of the mercury leveling bulb and the gas burette.

Hermetically sealed experimental chambers were used in some of the experiments. One type of these consisted of four-liter

[1] It should actually be 1 part of ethylene to 9,999 parts of air, but the difference is of course negligible and within the experimental error. For convenience of reference, this sort of difference will be disregarded in the descriptions and discussions which follow.

bottles supplied with large ground-glass stoppers. The bottles were calibrated approximately by pouring into them given volumes of water and marking the levels on the side of the bottle with a wax pencil. The bottles were then filled with water and the experimental materials were placed inside, after which the bottles were inverted in a pneumatic trough and the required volumes of the desired gases added by water displacement. The ground-glass stoppers were put in the bottles and sealed in place with sealing wax. This type of chamber was used only when the series of concentrations varied by increments of twenty-five per cent or more.

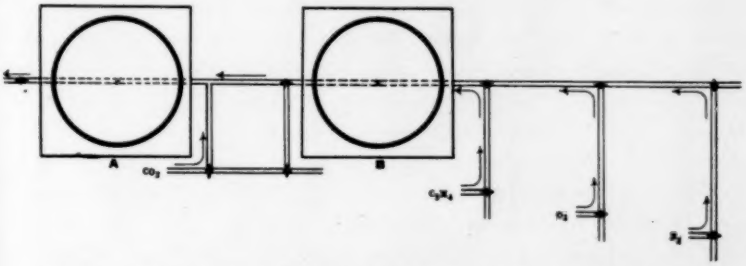


Fig. 2. Ground plan of cement bases, A and B, showing the arrangement of three-way stopcocks and glass tubes which make possible the addition or removal of known volumes of gases from any part of the system. The arrangement is figured as adjusted for experiments on exposure time, when cuttings in jar B were exposed to a mixture of ethylene, oxygen, and nitrogen in the absence of carbon dioxide, while those in jar A were exposed to the same gas mixture as in B with the addition of sufficient carbon dioxide to form a concentration of 10 per cent of the total gas volume.

It seemed desirable during the progress of this study to devise and construct an apparatus which would enable one not only to control the initial percentage composition of the experimental mixtures, but also to maintain this composition continuously by flowing the desired gases through the system. To meet these requirements an apparatus was constructed whereby gas mixtures of any desired composition or rate of flow could be passed continuously through hermetically sealed exposure chambers. The type of chambers used in this work was constructed as shown in figure 1. A base of cement, 40 cm. square and 8 cm. thick, was constructed with a groove in its upper surface which would just receive the rim of a twenty-

liter bell-jar. Inlet *a* and outlet *b* were embedded in the base at the time it was poured. The vertical portions of these delivery tubes were made of tin, since it was found to be nearly impossible to avoid breaking glass ones when removing or replacing the bell-jars. The bell-jar was placed upright on this base with its rim fitting into the groove, and a quantity of mercury sufficient to form a hermetical seal from 1–1.5 cm. deep was poured into the groove. Water was then added, so that a layer about .5 cm. deep covered the mercury. This was done to obviate possible toxic effects of mercury vapor. In some cases the bell-jars were tubulated at the top, which made possible the construction of the sealed opening (shown in detail at A in figure 1) that allows one to add or remove small objects without lifting the bell-jar from its base. Four chambers of the kind just described were connected in series, as shown in figure 2. The arrangement of glass tubes and three-way stopcocks allows one to add or remove volumes of gas at will from various parts of the system. All joints, whether glass to glass or tin to cement, were made with De Kotinsky's cement.

The gas mixtures which were passed through these enclosure chambers were controlled by either one of two methods. In the first method air was drawn or blown through the system by means of a vacuum-pressure pump which was connected to a water faucet. The water system used to run this pump was supplied with a constant-pressure reducing valve, next to the pump itself, in order to insure a more nearly constant rate of flow. This pump, as it normally functioned, drew about twice as much air through the vacuum side as it forced out of the pressure side, the other half being used in the pump in the formation of the pressure force. The rate of flow was maintained within about 10 per cent of the desired amount by regulation of the water pressure by the reducing valve, and by adjusting the pump itself.

The second method used for controlling the gas mixtures flowing through the chambers was by the use of the cylinders of compressed gases mentioned above. The cylinders were supplied with constant-pressure reducing valves which were connected with the inlets of the system of chambers (figure 2). It was possible to obtain a wide range of rates of flow from the cylinders by adjusting the relation between the size of the orifice

of the valve and the gas pressure back of it. The rates of flow were checked by determining, with a stopwatch, the length of time required to fill a gas burette or pipette with the gas. This same method was used at intervals to test the constancy of the rate of flow.

Adjustment of the relation between the size of the orifice and the pressure of the gas in the reducing valve controlled both the percentage composition and the rate of flow of the experimental gas mixtures through the chamber. Thus if the time periods required for 100 cc. of the gas to be liberated from the nitrogen and oxygen cylinders were, respectively, 45 seconds and 180 seconds, the resultant percentage composition of the experimental atmosphere would be 20 per cent of oxygen and 80 per cent nitrogen, while the rate of flow of these combined gases through the chamber would be 10 liters per hour.

Tests were made to determine the accuracy of this apparatus. These tests were made volumetrically by determining with a stopwatch the time required for a given volume of gas to be liberated into the system from each of the cylinders. In table 1 is given a representative set of such readings covering a period of six days during which an experiment was being made; the apparatus was not adjusted during that time. It is apparent

TABLE I

Stopwatch readings in seconds for period of six days of rate of flow of the four gases passing through the apparatus

KIND OF GAS	SIZE OF SAMPLE	TIME REQUIRED FOR LIBERATION OF SAMPLE					
		1st day	2nd day	3rd day	4th day	5th day	6th day
Nitrogen	100 cc.	45	44.9	46.2	46.5	47.2	49.
Oxygen	100 cc.	180	183	187	187	187	188
Carbon dioxide	100 cc.	361	375	365	369	385	385
Ethylene	10 cc.	542	570	595	566	566	568

from an examination of the data that the rate of flow of each gas was maintained with a range of variation not exceeding one-tenth of the desired concentration. An analysis with an Orsat apparatus of the experimental atmosphere as it came from the exposure chambers showed a similar degree of accuracy of control.

DESCRIPTION OF RESULTS

The general character of the intumescences

It is obvious from a review of the literature as given above that various pathological outgrowths on roots, stems, and leaves which consist chiefly of hypertrophy of the affected tissues have been termed "intumescences." More careful cytological and experimental studies of these outgrowths may make possible a more adequate classification of them. The proposal of Küster (1903) to limit the term intumescence to small pustule-like overgrowths is apparently not based on any other than artificial considerations of convenience. Both Sorauer (1886) and Küster (1903) have used the term to include a wide range of overgrowths varying as to the relative degree of hyperplasia and hypertrophy involved, the characters of the tissues produced, and the stimuli which lead to their development. Küster (1903) notes that such "intumescences" do not differ in their method of development from the more extensive lenticel overgrowths for which he used the term excrescence.

As I reported in my preliminary paper, when conditions are favorable intumescences due to ethylene stimulation may appear in the buds, in the proximal or distal ends, and in the internodal regions of stem cuttings of Transparent apple (see Wallace, 1926, plates 14 and 15, fig. 1–11). As a rule the buds first show intumescences. The reaction appears first on the surface as a transverse breaking of the epidermis and cork layers on each side of a bud base. The time required for the appearance of this response may be as short as forty-eight hours or as long as six days. Four days, however, is usually sufficient to bring about the response. Proliferation and maceration of the cells of the bud and bud base may proceed rapidly, and within a few days the entire resting bud containing the leaf primordia for the following year may be resolved into a loose mass of rounded cells. Externally the buds in this condition present an appearance very suggestive of popped corn. The curled remnants of bud scales, cork layers, etc., adhere to the enlarging tissues beneath. This swollen, white tissue is so friable that a slight jar often suffices to cause most of the intumescence to fall from the stem. This may, in some cases, result in the exposure of tissue as deep-seated as the xylem of the stem. Not infrequently

such an intumescence may occur in the region of the bud-scale scars which mark the position of the terminal buds of previous years.

Intumescences in the distal ends of the cuttings begin before the bud intumescences have reached their complete development (that is, in from five to eight days after exposure to the gas), and their development is similar to that described for buds. The first evidence of intense responses, in the case of twigs cut off at both the basal and the apical ends, is a swelling that produces a longitudinal rupturing of the epidermis and cork layers near the apical end. Swellings of this kind may extend back from the tips of the cuttings for 2–3 cm. in some cases. Proliferation and swelling then proceed as in the region of the buds, but the disorganization of the tissues is never so complete. Frequently, most of the cell enlargement is limited to the deeper regions of the cortex, with the result that a layer composed of the outer layers of the phloem and cortex, containing the chlorenchyma, is split off and raised upon a pad of loose parenchyma. In all cases, the chlorenchyma layer retains its green color, though it may be torn so as to expose rifts or lesions lined by light colored, hypertrophied cells. The zone of transition between the swollen region of the tip and the normal regions below is generally rather sharp. Two or even three sharply delineated bands of swollen tissue similar to those which form in the region of the tips may be present on one cutting, distributed at irregular intervals along its length. The tissues lying between these swollen zones are apparently normal. The increase in the diameter of the stem in the region of a swollen zone usually varies directly with the width of the zone. Swollen zones that are very narrow may raise the bark only 1 mm. or less, while those which are 2 cm. or more in width may completely rupture the epidermis and cork layers and increase the diameter of a twig whose normal diameter is only 4 mm. to as much as 6–8 mm.

Intumescences in the internodal regions occur later than those in the ends and appear in from nine to twelve days after exposure to the gas; they reach their maximum development five to ten days later. These swellings may be relatively superficial, or they may form deep-seated cankers that penetrate to the xylem by long, deep, longitudinal rifts that run the entire

length of the cutting. In a superficial type of response (plate 34 figure 1), the phellogen proliferates so as to produce a splitting and rolling back of the cork, thus exposing extensive areas of the chlorenchyma. Examination of the surface of the chlorenchyma of a cutting of this kind shows it to be made up of large, globular, hypertrophied cells. Other cuttings may show, in addition to proliferation of the phellogen and hypertrophy of the chlorenchyma, one or more longitudinal rifts which penetrate into the deeper layers of the bark. The most deepseated type of intumescence is well shown in plate 34, figure 2. Here the disorganization of the tissues in certain areas is almost as complete as that found in the proliferated buds. In general the cuttings which I have studied were affected about equally throughout their entire length, but in some cases there seemed to be a tendency for the lower portions to react less strongly or not at all. Usually in cases where an internodal swelling is situated below a terminal swelling on the same cutting, the two tend to run together.

The formation of intumescences successively in the buds, in the internodal, and in the distal regions seems to be the typical response of the cuttings of Transparent apple to stimulation by ethylene gas.[2]

Populus deltoides, when exposed to ethylene gas, forms intumescences at the base of the buds or in the internodes, but no swellings have been observed at the cut ends.[3] The internodal responses are rarely conspicuous and frequently may not occur at all, but large intumescences generally appear at the base of the buds. The response usually begins within about five days after the cuttings have been exposed to the gas. Proliferation at the base of the buds gives rise to a pad of soft, white tissue that completely separates the bud from the stem, though, in contrast with the case of apple, the leaf primordia usually

[2] Three varieties of apple which I have studied indicate that there may be a great varietal variation in the capacity to form these outgrowths. Transparent has always shown the greatest capacity for response. A large tree, of unknown variety, formed intumescences in the three regions, but they were never strong, and were often lacking. A third variety, Williams, never formed an intumescence in the buds or in the internodes, but did form strong intumescences in the distal ends.

[3] These intumescences have formed equally well in concentrations of 1 per cent ethylene and 1 part of ethylene to 10,000 parts of air.

remain intact. The cells of this mass of tissue are long and sac-like and cling together to form a delicate cotton-like mass; they are not loose and granular like those in the buds of apple. At the end of a week such an intumescence may be 1 mm. in diameter. By the end of a month it may be as much as 1 cm. long by 0.8 cm. wide and 0.5 cm. thick (Plate 34, figures 3, 4, and 5). I have not determined the maximum size which such an outgrowth may attain.

The hypertrophied cells which make up the greater part of these intumescences in apple are snow-white in mass, but are almost devoid of color when viewed as individual units. They vary from globular to ovoid or sometimes almost filamentous in form. The cell walls are always extremely thin. The nuclei appear normal, but the plastids aggregate in a thickened band of the primordial utricle around the equator of the cell. Typically the plastids are arranged in one such band, but not infrequently a single cell may contain two or even three bands placed at irregular intervals between the two extremities of the cell, but not necessarily parallel to each other. Starch grains, which are very conspicuous in the plastids of the hypertrophied cells at the time of their formation, gradually disappear as the cells grow older, so that none are present after three or four weeks. The cells then die and turn brown, apparently through exhaustion of their food reserves.

My experiments show that the intumescences of apple cuttings described above may form under a wide range of environmental conditions. They were obtained in stems ranging in age from young succulent growths of the current year to stems as much as twelve years old.[4] Likewise, cuttings which have been kept as controls for a month have later given the response after being exposed to the gas. Preliminary experiments indicate that acetylene, the next member of the unsaturated hydrocarbon series, may induce a similar type of response.

The following experiments were made, unless stated to the contrary, on cuttings of Transparent apple. The pieces used were 10–15 cm. long and of graded sizes from 1.5 cm. in diameter

[4] Intumescences similar to those formed in the internodes of the stems have been formed on the large and small (1 cm.–5 cm.) roots collected from the same trees. These roots were probably of some other variety than Transparent since the trees were grafted stock.

down to slender terminal twigs. The data recorded in the tables represent the maximum response which the cuttings showed under the conditions of the experiment. The length of time required for maximum development of the intumescences varied from about a week for those at the base of buds to two or more weeks for those at the ends of the cuttings and in the internodes. The degree of development of these swellings is recorded in the tables as: none, weak, medium, strong, and maximum. I report no special experiments on the effect of light because from the very first it was evident that the reactions took place equally well whether in complete darkness, in continuous light, or in alternate light and darkness.

TABLE 2

Response to ethylene in different branches from the same tree[1]

BRANCH NO.	POSITION OF BRANCH	NO. OF PIECES	LOCATION OF INTUMESCENCES					
			Internodes		Distal ends		Buds	
			Degree	No.	Degree	No.	Degree	No.
1	Top of the tree	8	strong	7	weak	6	medium	6
2	Second from top	8	control		none		none	
3	Third from top	8	maximum	8	weak	7	medium	17
4	Fourth from top	8	strong	6	medium	6	medium	8
5	Middle of tree	8	maximum	8	weak	3	medium	8
6	Middle of tree	8	strong	7	medium	7	medium	8
7	Middle of tree	8	strong	8	medium	7	medium	8
8	Base of the tree	8	strong	8	medium	7	medium	14

[1] The character of the responses is graded as follows: none, weak, medium, strong, and maximum. The numbers given for the internodal and apical responses refer to the number of cuttings showing the response; those for the bud responses refer to the number of buds reacting and not to the number of stems on which the buds were borne.

Tests begun July 14; concentration of ethylene, 1 part to 40 parts of air; exposure time, 48 hours.

General preliminary experiments

Experiments made on branches taken from various parts of the same tree showed no significant variation in the sensitiveness of different branches to stimulation by ethylene gas. Eight branches were collected at one time from different parts of the same tree and cut into eight cuttings each. These sets were then placed in 15-liter bell-jars (one set to a jar), and a volume

of ethylene sufficient to produce a concentration of 1 part of ethylene to 40 parts of air was liberated into each bell-jar by

TABLE 3

Response to ethylene in branches from different trees of the same variety of apple[1]

TEST NO.	DATE	NO. OF TREE	NO. OF PIECES	LOCATION OF INTUMESCENCES						ABSCISSION OF LEAVES	
				Internodes		Distal ends		Buds			
				Degree	No.	Degree	No.	Degree	No.	Degree	No.
1	July 18	1	8	weak	5					complete	2
		2	8	none						complete	2
		3	8	weak	6					complete	1
		4	8	weak	1					partial	
		5	8	weak	2					partial	
		6	8	none						partial	
		7	8	none						partial	
		8	8	none						partial	
2	July 21	1	8	medium	1	medium	3	weak	4	complete	5
		2	8	medium	1	medium	3	medium	2	complete	7
		3	8	none		weak	2	none		complete	5
		4	8	weak	2	medium	3	none		complete	7
		5	8	weak	1	weak	1	none		complete	7
		6	8	weak	1	medium	2	medium	3	complete	4
		7	8	weak	1	none		none		complete	7
		8	8	medium	1	medium	2	medium	2	complete	5
3	July 27	1	8	strong	4	medium	4	medium	4	partial	
		2	8	strong	4	medium	4	medium	3	complete	6
		3	8	none		strong	5	weak	2	complete	3
		4	8	strong	8	strong	8	strong	10	complete	8
		5	8	none		medium	3	none		partial	
		6	8	weak	3	medium	4	none		complete	8
		7	8	medium	3	medium	7	strong	3	complete	8
		8	8	weak	2	medium	4	medium	5	complete	8

[1] Tests 1 and 2 were made on cuttings from the same eight trees, while test 3 was made from cuttings from eight other trees of the same variety. The degree of abscission of leaves is designated as partial when only part of the leaves fell from a stem, and complete when all fell.

Concentration of ethylene, 1 part to 40 of air; exposure time, 48 hours.

removing the cover from a small vessel containing ethylene which had been inclosed in the large bell-jar, as described above.

The jars were then left closed for forty-eight hours, after which the ground-glass plates were removed for a like period of time and then replaced. The jars then remained closed for the remainder of the experiment except during the brief periods in which readings were being made. Cuttings from every branch, whether from the top or the base of the tree, developed intumescences in the buds and internodes and at the distal ends (table 2).

Similar experiments with branches taken from eight different trees of Transparent failed to show a variation great enough to indicate any essential difference in their susceptibility. The variation in the number of pieces showing intumescences and the degree of the response, as shown in table 3, were of the same order of magnitude as the variations in pieces from the same branch.

TABLE 4

Response to ethylene in cuttings collected at different times of the year[1]

TEST NO.	DATE	NO. OF PIECES	LOCATION OF INTUMESCENCES					
			Internodes		Distal ends		Buds	
			Degree	No	Degree	No.	Degree	No.
1	Oct. 29	6	medium	4	medium	4	none	
2	Nov. 10	12	none		strong	12	none	
3	Nov. 16	10	none		medium	9	medium	3
4	Nov. 24	8	none		medium	7	none	
5	Dec. 21	15	none		none		none	
6	Jan. 1	15	maximum	15	maximum	15	maximum	15
7	Jan. 5	15	medium	15	medium	15	medium	2
8	Jan. 18	15	none		none		none	
9	Feb. 1	5	none		weak	1	weak	1
10	Feb. 7	5	none		none		none	
11	Feb. 14	5	medium	4	medium	4	medium	3
12	Feb. 23	5	none		medium	2	none	
13	Mar. 9	5	none		none		none	
14	Mar. 22	5	none		none		none	
15	Mar. 29	5	weak	1	none		none	
16	Apr. 4	5	medium	1	none		medium	3
17	Apr. 12	5	weak	4	none		none	
18	Apr. 21	5	weak	4	none		none	
19	Apr. 30	5	none		medium	1	weak	2
20	May 12	5	none		none		none	
21	June 3	5	none		weak	3	none	

[1]Concentration of ethylene, 1 part to 200 parts of air; exposure time, varied,—usually 48 hours.

I reported in my preliminary paper that such intumescences could be produced during the fall and winter period from September to March, and I can now report like results for the entire year. In table 4 are given the results for a series of tests extending from October, 1925, to June, 1926. As will be seen from the table there was great variability in the results of these experiments during this period. In another series treatments of new lots of cuttings were started four times daily for a period of twelve days. The results given in table 5 for this short interval show the same irregularities as those shown in table 4 for similar experiments extending over a period of eight months. These two series indicate that the irregularities in the responses secured were not due to differences in the internal condition of the twigs that might have been correlated with their seasonal condition when collected. These experiments were made under

TABLE 5

Response to ethylene in cuttings made four times daily for twelve days[1]

TEST NO.	TIME[2]		NO. OF PIECES	LOCATION OF INTUMESCENCES					
				Internodes		Distal ends		Buds	
	Day	Hour		Degree	No.	Degree	No.	Degree	No.
1	7	12 n.	6	maximum	4	medium	4	weak	3
2	7	6 p.m.	7	medium	2	medium	6	weak	2
3	7	12 m.	8	medium	4	medium	6	weak	3
4	8	8 a.m.	5	weak	3	medium	4	weak	4
5	8	12 n.	8	medium	3	medium	7	medium	4
6	8	6 p.m.	7	medium	4	medium	5	medium	5
7	8	12 m.	11	strong	6	weak	6	medium	4
8	9	8 a.m.	7	weak	2	weak	5	medium	15
9	9	12 n.	10	weak	7	weak	5	medium	7
10	9	6 p.m.	7	weak	2	medium	4	none	
11	9	12 m.	9	weak	2	weak	3	weak	1
12	10	8 a.m.	6	weak	1	medium	2	weak	1
13	10	12 n.	11	medium	4	weak	6	weak	2
14	10	6 p.m.	9	weak	7	medium	5	weak	1
15	10	12 m.	12	medium	7	weak	6	medium	9
16	11	8 a.m.	7	medium	4	weak	4	medium	6
17	11	12 n.	10	weak	6	strong	7	medium	4
18	11	6 p.m.	10	medium	3	medium	2	weak	1
19	11	12 m.	11	weak	1	weak	1	weak	2
20	12	8 a.m.	6	weak	1	weak	2	none	
21	12	12 n.	9	weak	3	none		none	

TABLE 5—*Continued*

TEST NO.	TIME[2]		NO. OF PIECES	LOCATION OF INTUMESCENCES					
				Internodes		Distal ends		Buds	
	day	Hour		Degree	No.	Degree	No.	Degree	No.
22	12	6 p.m.	10	weak	2	weak	2	none	
23	12	12 m.	12	weak	3	weak	1	none	
24	13	8 a.m.	0						
25	13	12 n.	12	weak	1	none		none	
26	13	6 p.m.	11	none		none		strong	1
27	13	12 m.	9	none		weak	4	none	
28	14	8 a.m.	6	none		medium	1	none	
29	14	12 n.	10	none		weak	1	none	
30	14	6 p.m.	12	none		weak	2	none	
31	14	12 m.	11	none		weak	1	none	
32	15	8 a.m.	5	none		medium	2	none	
33	15	12 n.	9	none		none		none	
34	15	6 p.m.	11	none		none		none	
35	15	12 m.	9	none		none		none	
36	16	8 a.m.	11	weak	2	strong	1	medium	11
37	16	12 n.	8	maximum	7	weak	2	maximum	35
38	16	6 p.m.	6	medium	5	weak	4	medium	11
39	16	12 m.	6	weak	4	medium	4	weak	5
40	17	8 a.m.	6	weak	3	weak	2	weak	1
41	17	12 n.	6	weak	3	none		none	
42	17	6 p.m.	6	none		none		none	
43	17	12 m.	6	none		none		none	
44	18	8 a.m.	6	weak	3	weak	3	medium	6
45	18	12 n.	6	none		none		none	
46	18	6 p.m.	6	none		none		none	
47	18	12 m.	7	none		none		none	
48	19	8 a.m.	8	none		none		none	

[1] Concentration of ethylene, 1 part to 40 of air; exposure time, 48 hours.

[2] The tests were made during June, 1926; the calendar days of the month are shown. Noon is denoted by n., midnight by m.

greenhouse conditions. The temperature fluctuated through a range from 15° to 40° C. Special experiments planned to test the influence of temperature indicate that high temperatures were probably responsible for the failure to get responses in certain cases given in tables 4 and 5. In the experiments conducted from June, 1926, to March, 1927, care was taken to avoid excessively high or low temperatures, and consistant responses were obtained in every test of the entire series.

Effect of varying environmental conditions on the response to ethylene

Relation of humidity to response. Varying degrees of atmospheric moisture, within the range studied, were found to have little or no influence upon the reaction of apple twigs to ethylene. The humidity was controlled in a series of hermetically sealed bell-jars, as described above. Two of the bell-jars were connected to the vacuum side and two to the pressure side of the vacuum-pressure pump. In this manner air could be drawn through the first two bell-jars at a rate of about 40 liters per hour and then passed through the pump and forced back through the other two bell-jars at a rate of about 20 liters per hour. Calcium chloride tubes were arranged between the bell-jars so as to maintain relative humidities of 67, 75, 84, 88, and 92 per cent. Actual tests of the relative humidity of the air in each chamber were made three times daily with a wet and dry bulb hygrometer (hygrodeik), and the average values of these readings for the period of seven days during which the experiment was continued are recorded in table 6. The five sets of cuttings were exposed to 1 part of ethylene to 100 of air for twelve hours prior to being transferred to the bell-jars with controlled humidity. The gas mixture in the exposure chamber

TABLE 6

Relation of humidity to the formation of intumescences[1]

RELATIVE HUMIDITY	NO. OF PIECES	INTUMESCENCES IN THE DISTAL ENDS	
		Degree	No.
per cent			
67	10	weak	9
75	10	strong	5
84	10	medium	6
88	10	maximum	8
92	10	strong	8

[1]Tests begun Oct. 25: Concentration of ethylene, 1 part to 100 of air; exposure time, 12 hours.

was made up of volume proportions of 1 part of ethylene, to 20 parts of oxygen, to 80 parts of nitrogen, in addition to the water vapor needed to give the desired humidity; and these proportions were maintained during the period of exposure to

the ethylene by regulating the rate of liberation of the three gases from cylinders by means of pressure-reducing valves, as described above. The basal end of each cutting was in contact with either moist sand or moistened filter paper, as in all the other experiments. The temperature, and hence the water vapor pressure deficit, remained nearly constant during the experiment. As shown in table 6, intumescences formed on cuttings in all the gas chambers. With a humidity of 67 per cent the response was weak, but nine out of ten cuttings reacted. In the jar with a humidity of 84 per cent the intumescences were fairly well developed, but occurred on only six of the ten cuttings. Strong responses occurred on five cuttings in the jar with a humidity of 75 per cent, and on eight cuttings in the jar with a humidity of 92 per cent. It would seem, therefore, that relative humidity within the range of 67–92 per cent had little or no influence on the response.

Relation of temperature to response. Experiments made with temperatures ranging from about 15° C. to 30° C. showed definite effects on the degree of the response, but neither extreme exceeded the range in which intumescences may form. Four twelve-liter bell-jars were placed so as to maintain various temperatures. One container was put on a laboratory table where the room temperature was about 20° C. Another was placed in an ice-box with a temperature of about 15° C. The temperature values given in table 7 for these two jars are the averages of their readings for the period of the experiment. The other two containers were kept in electric incubators adjusted to maintain temperatures of 25° and 30° C., respectively. These containers were filled with 1 per cent of ethylene and 99 per cent air, and sets of 8–14 cuttings were placed in each. Some intumescences developed at each temperature, but there were pronounced differences in the intensity of the responses at the different temperatures. In these tests no internodal swelling formed on any of the twenty-nine cuttings kept at 30° C. Weak to medium terminal intumescences developed on 12 of them. At 25° C. weak to medium internodal swellings formed on sixteen cuttings out of twenty-nine, while twenty of the twenty-nine showed medium responses at their distal ends. Responses at room temperatures (19–20.5° C.) were much stronger than those at the two higher temperatures; since medium to weak terminal

TABLE 7

Relation of temperature to the formation of intumescences[1]

TEST NO.	DATE	AVERAGE TEMPERATURE C.	NO. OF PIECES	LOCATION OF INTUMESCENCES			
				Internodes		Distal ends	
				Degree	No.	Degree	No.
1	Nov. 24	15.0	13	maximum	12	maximum	12
		20.5	14	medium	12	medium	12
		25.0	13	medium	10	medium	10
		30.0	12	none		weak	8
2	Dec. 1	15.0	8	strong	7	strong	7
		19.0	8	medium	4	medium	4
		25.0	8	weak	6	medium	6
		30.0	8	none		medium	1
3	Dec. 8	14.8	8	maximum	7	maximum	7
		19.5	8	weak	6	weak	6
		25.0	8	none		medium	6
		30.0	8	none		weak	3

[1]Concentration of ethylene, 1 part to 100 parts air; exposure time, throughout the experiment.

and internodal swellings formed on twenty-two out of the thirty cuttings. The most pronounced responses were obtained at 15° C., the lowest temperature of the entire series. These intumescences were among the best developed that I have had. They involved almost the entire length of the cuttings and showed deep rifts and high ridges of proliferated tissue. Also, as will be observed from table 7, the percentage of cuttings responding at 15° C. was very high, only three of the twenty-nine failing to develop intumescences.

Conditions affecting penetration by the gas. My experiments have shown apparently that many buds of Transparent apple may fail to form intumescences because the ethylene has not penetrated into them. Usually only one bud in ten to fifteen gives a response after the cuttings have been exposed to ethylene. I have been able, however, to obtain responses in about 90 per cent of the buds by cutting away a portion of the thickened, corky leaf-scar at the base of each bud. Table 8 gives the results obtained in a representative experiment. Only three buds out of a total of fifty-nine developed intumescences when

TABLE 8

Effect on intumescence formation of removal of the abscission layer at base of buds[1]

CUTTING NO.	ABSCISSION LAYER NOT REMOVED		ABSCISSION LAYER REMOVED	
	No. of buds	No. of responses	No. of buds	No. of responses
1	5	1	2	1
2	3	0	2	2
3	2	0	1	1
4	3	0	6	6
5	5	1	2	2
6	3	0	10	10
7	3	0	3	3
8	6	0	8	8
9	9	0	3	1
10	6	0	2	2
11	4	1	2	2
12	7	0	2	2
13	3	0	5	4
14			4	3
15			6	6
Totals	59	3	58	53

[1]Concentration of ethylene, 1 part to 100 of air; exposure time, throughout the experiment.

the corky layers were not removed, while of fifty-eight other buds from whose bases the corky layers were removed, only 5 failed to respond. Moreover, after the layers had been removed from thirty-two of the buds mentioned above which had failed to respond to ethylene, all but 5 gave responses. However, this removal of the corky layer did not always assure a response and in some cases nearly every bud on a cutting has given a response without removal of the corky layers. Like attempts to induce responses in the buds of Williams, a variety which has never given bud responses, by cutting the base of the buds proved unsuccessful.

In order to study the influence of facilitating the access of the ethylene to the tissues, the corky layers of the bark were removed in some of the tests. This treatment did not always result in more extensive intumescences than those produced on cuttings with uninjured bark. Pieces of the living bark that were stripped from the stems and exposed to ethylene showed

little or no proliferation. Further study will be required before definite statements can be made as to the part that lenticels and various types of wounds may play in affecting the absorption of ethylene by twigs.

Dosage effects. The most striking fact regarding the formation of these intumescences on the stems of Transparent apple is perhaps the constancy of the results under extreme ranges of concentration of the gas. Mixtures composed of 75 per cent of ethylene and 25 per cent of oxygen give about the same response as others made up of 1 part of ethylene to 10,000 parts of air. There is an apparent decrease in the intensity of the response when the concentration is changed from 1 part of ethylene and 10,000 parts of air, to 1 part of ethylene and 1,000,000 parts of air. Still, definite intumescences have formed on the distal ends of apple cuttings when treated with dilutions of 1 part to 1,000,000, and 1 part to 100,000,000. The reactions in these two last concentrations are always weak and are slower in making their appearance, but they are distinct.

Numerous experiments have shown that intumescences may form on the stems of Transparent apple in response to extremely low concentrations of ethylene gas. Thus, as shown in test 1 table 9, a series of concentrations ranging from 75 per cent ethylene and 25 per cent oxygen, to 1 part of ethylene to 10,000 parts of air gave well developed intumescences. Within this range of concentrations the intensity of the response seems to bear little or no relation to the gas concentration, as is shown by both qualitative and quantitative measurements of the reaction. An examination of the data of the five tests likewise indicates that there was no significant decrease in the intensity of the response when the concentration of ethylene was reduced from 1 per cent to 1 part of ethylene to 10,000 parts of air. However, a further decrease in the concentration, from 1 part to 10,000 to 1 part to 1,000,000 or 100,000,000, showed a definite decrease in the intensity of response. The intumescences formed in these last two concentrations were always on the ends of the cuttings and usually extended back from the tips only a few millimeters, but they were always distinct.

The interval of time necessary for the appearance of the responses was directly proportional to the concentration of

TABLE 9

Relation of extreme ranges of concentrations of ethylene to the formation of intumescences[1]

TEST NO.	DATE	NO. OF PIECES	COMPOSITION OF MIXTURE		COMBINED INTUMESCENCES IN INTERNODES AND DISTAL ENDS		
			Air	Ethylene	Qualitative		Quantitative Av. length in cm.
					Degree	No.	
1	Nov. 15	8	25[2]	75	maximum	5	15.0
		7	75	25	maximum	6	12.0
		6	100	1	maximum	6	8.5
		7	10,000	1	maximum	6	16.0
2	Nov. 19	13	100	1	maximum	13	15.4
		11	10,000	1	maximum	10	13.0
		11	1,000,000	1	medium	10	5.9
		11	100,000,000	1	weak	9	4.1
3	Dec. 1	8	100	1	medium	5	
		8	10,000	1	medium	7	
		8	1,000,000	1	weak	4	
		8	100,000,000	1	weak	4	
4	Feb. 14	10	100	1	maximum	9	7.7
		10	10,000	1	maximum	9	6.5
		10	1,000,000	1	weak	3	.5
		10	100,000,000	1	weak	2	.2
		10	control	0	none		
5	Mar. 15	10	100	1	strong	5	3.2
		10	10,000	1	strong	3	4.8
		10	1,000,000	1	medium	5	.9
		10	100,000,000	1	medium	3	.6
		10	control	0	none		

[1]Exposure time: test 1, 72 hours; tests 2 and 3, 1 week; tests 4 and 5, two weeks.

[2]This one combination was oxygen-ethylene, only; all others were ethylene-air, as shown.

ethylene, in these tests with series of concentrations from 1 per cent to 1 part to 100,000,000. The terminal responses usually appeared after three to four days in the highest concentration. The responses in the other concentrations then followed at intervals of 24 to 48 hours, following the descending series

of concentrations. Thus, it was usually about a week before any response was evident in the mixture of 1 part of ethylene to 100,000,000 parts of air.

The methods of controlling the ethylene concentration and of conducting the experiments were varied in order to insure accuracy in the composition of the mixture and to check the constancy of the results. The gas chambers for tests 1, 2, and 3 (table 9) were twenty-liter bell-jars, hermetically sealed, in the manner described above, by means of cement bases and mercury. Those for tests 4 and 5 were similar jars inverted and sealed by means of ground-glass plates and castor oil. Gas mixtures in which the concentration of ethylene was greater than 1 per cent were made by flowing the ethylene and other gases through the jars in the required proportions during the entire exposure period for the experiment. All concentrations of ethylene from 1 per cent to 1 part to 100,000,000 of air were made with the gas burette and leveling bulb, as described. The concentrations were made in an ascending series in order that the amount of ethylene which might remain in the burette, when changing from one concentration to the next higher, would be insignificant when compared with the amount in the mixture of higher concentration being made. Exposure periods were seventy-two hours for test one, one week for tests two and three, and two weeks for tests four and five. In no case was there renewal of the gas mixture inside a chamber during the period of a test.

Relative effects of illuminating gas and ethylene. Ethylene and illuminating gas have given about the same responses over a wide range of concentrations. Sets of cuttings were placed in twenty-liter bell-jars containing air and quantities of ethylene sufficient to form concentrations of 0.05, 0.5, 5, 25, 50, and 100 per cent (table 10). Similar preparations were made with illuminating gas and air so as to obtain parallel series of concentrations of the two gases. The jars containing 100 per cent of ethylene or illuminating gas were filled by water displacement through tubulations at the top and bottom of the jars. All other concentrations were made by liberating ethylene or illuminating gas from vessels which had been inclosed in the inverted bell-jars filled with air and containing the cuttings. The jars were then left closed for 48 hours for the exposure

of the cuttings to the gas mixtures, after which they were left open for a like period of time. They then remained sealed for the remainder of the experiment except for the brief intervals of time in which readings were being made. The containers for most of these experiments were inverted bell-jars, as described above; but a like series of experiments made in hermetically sealed four-liter bottles gave essentially the same results.

Although the tests of the ethylene series gave responses with all concentrations except that of pure ethylene, the reaction in the lowest concentration was always weak. The illuminating gas series failed to give intumescences with both the lowest and the highest concentrations. No definite differences in the intensity of the responses for the intermediate concentrations of the two gases were observable. It might be concluded from these experiments that the lowest concentration of the illuminating gas contained an amount of ethylene that was not sufficient to give a response, and that the same concentration of pure ethylene was sufficient to induce only a weak response. This may be correct for the tests with the lowest concentration of the illuminating gas; but, as has been shown above, ethylene

TABLE 10

Effect of various parallel concentrations of ethylene and illuminating gas[1]

TEST NO	DATE	CONCENTRATION OF GAS MIXTURE		NO. OF PIECES	LOCATION OF INTUMESCENCES					
					Internodes		Distal ends		Buds	
		Ethylene	Illuminating gas		Degree	No.	Degree	No.	Degree	No.
		per cent	*per cent*							
		.05		6	weak	2	none		weak	1
		.5		6	weak	4	medium	4	weak	3
		5.		6	medium	5	weak	2	medium	1
		25.		6	weak	3	medium	5	medium	4
		50.		6	medium	3	strong	3	medium	1
		100.		6	none		none		none	
1	June 28		.05	6	none		none		none	
			.5	6	weak	1	weak	1	weak	1
			5.	6	weak	3	weak	2	none	
			25.	6	weak	3	weak	4	none	
			50.	6	weak	2	medium	4	none	
			100.	6	none		none		none	

TABLE 10—*Continued*

TEST NO.	DATE	CONCENTRATION OF GAS MIXTURE		NO OF PIECES	LOCATION OF INTUMESCENCES					
					Internodes		Distal ends		Buds	
		Ethylene	Illuminating gas		Degree	No.	Degree	No.	Degree	No.
		.05		5	weak	1	weak	1	none	
		.5		5	medium	4	medium	1	strong	5
		5.		5	strong	4	strong	2	maximum	10
		25.		5	medium	5	medium	3	medium	5
		50.		5	strong	4	weak	2	weak	6
		100.		5	none		none		none	
2	July 8									
			.05	5	none		none		none	
			.5	5	medium	5	strong	3	weak	5
			5.	5	strong	4	medium	3	strong	4
			25.	5	medium	5	strong	3	medium	8
			50.	5	medium	5	medium	4	medium	5
			100.	5	none		none		none	
		.05		6	weak	2	none		weak	1
		.5		6	weak	4	medium	4	none	
		5.		6	medium	5	weak	2	none	
		25.		6	weak	3	strong	5	medium	4
		50.		6	medium	3	strong	3	strong	1
		100.		6	none		none		none	
3	June 20									
			.05	6	none		none		none	
			.5	6	weak	1	weak	1	none	
			5.	6	weak	3	weak	2	none	
			25.	6	weak	3	weak	4	weak	1
			50.	6	weak	2	strong	4	none	
			100.	6	none		none		none	
		.05		5	weak	1	none		none	
		.5		5	medium	3	strong	5	strong	4
		5.		5	strong	4	medium	2	strong	11
		25.		5	medium	5	strong	4	weak	4
		50.		5	strong	2	strong	4	weak	3
		100.		5	none		none		none	
4	July 13									
			.05	5	none		none		none	
			.5	5	none		strong	5	medium	3
			5.	5	strong	5	medium	4	medium	6
			25.	5	strong	3	strong	3	strong	1
			50.	5	medium	2	strong	3	none	
			100.	5	none		none		none	

[1]Exposure time, 48 hours.

has often given strong responses in concentrations much lower than 1 part to 2,000 (0.05 per cent).

The highest concentration of ethylene (100 per cent) did not apparently kill the cuttings nor did it produce observable injury. The highest concentration of illuminating gas (100 per cent), on the other hand, always caused obvious injury and not infrequently killed the cuttings. The higher toxicity of illuminating gas as compared with pure ethylene was unexpected, since other investigators have frequently regarded ethylene as the most toxic constituent for plants of the illuminating gas which they have used.

Effect of mixtures of ethylene with oxygen, nitrogen, and carbon dioxide. A series of experiments with mixtures of oxygen, nitrogen, carbon dioxide, and ethylene, in which the constituents were varied in their relative proportions by increments of 25 per cent, have shown that intumescences may fail to form in the absence of oxygen or in the presence of 25 per cent or more of carbon dioxide. This series of mixtures was made by water displacement, according to the combinations and percentages shown in table 11. When ethylene was not one of the primary constituent gases of the experimental atmosphere, it was added so as to be present as 1 part to 20 parts of the other gases. As shown by the nitrogen-oxygen and oxygen-ethylene series, a percentage of 25 per cent or more of oxygen seemed to have little influence in increasing the intensity of the response. That oxygen is necessary for the occurrence of the response is well shown, however, since no proliferation or swelling of any kind occurred in its absence. The inhibiting influence of 25 per cent or more of carbon dioxide was very pronounced. Such a concentration of carbon dioxide prevented completely the formation of any intumescences even in cases where the oxygen content was four times that of ordinary air.

Relation of exposure time to response. A series of experiments was made to determine the effect of varying the exposure time on the type and character of the intumescences formed. In this series the gas mixture consisted of 1.5 per cent ethylene amd 98.5 per cent air. The exposure time was varied so as to form a series ranging from 7½ minutes to the entire period of the experiment (about two weeks). As shown in table 12, intumescences formed in every case, and there was no apparent

TABLE II

Effect of various gas mixtures on formation of intumescences and abscission of leaves[1]

CONCENTRATION IN GAS MIXTURE				LOCATION OF INTUMESCENCES				ABSCISSION OF LEAVES	
				Internodes		Buds			
Nitrogen	Oxygen	Carbon dioxide	Ethylene	Degree	No.	Degree	No.	Degree	No
per cent	*per cent*	*per cent*	*per cent*						
100	0			none		none		none	
75	25		[2]	strong	3	weak	1	complete	4
50	50			weak	3	strong	4	complete	5
0	75			weak	3	strong	10	complete	5
	100	0		medium	3	medium	3	complete	5
	75	25		none		none		none	
	50	50		none		none		none	
	25	75		none		none		none	
	0	100		none		none		none	
25			75	none		none		none	
50			50	none		none		none	
75			25	none		none		none	
	75		25	medium	3	strong	5	complete	5
	50		50	weak	3	strong	5	complete	5
	25		75	strong	2	maximum	4	complete	3
	0		100	none		none		none	

[1] Test begun July 22; exposure time, throughout the experiment; number of cuttings, five in each set.

[2] When ethylene was not one of the primary constituent gases of the mixture it was added as one part to twenty to the completed mixture.

correlation between length of the exposure time and the intensity of the reaction. In view of the extremely low concentration of ethylene which is sufficient to give a response after an exposure time of one to three days, it is not surprising to find that a concentration of 1.5 per cent gives a response after an exposure time of only 7½ minutes.

GENERAL DISCUSSION

The results reported show that intumescences may develop in the stems of apple in response to stimulation by ethylene gas under a wide range of conditions. My results are somewhat similar to those described by Von Schrenk for the intumescences formed on the leaves of cauliflower in response to stimulation

TABLE 12

Relation of exposure time to formation of intumescences[1]

TEST NO.	DATE	EXPOSURE TIME	NO. OF PIECES	INTUMESCENCES IN DISTAL ENDS OF CUTTINGS	
				Degree	No.
1	Oct. 15	7½ min.	5	weak	4
		15 min.	5	weak	3
		30 min.	5	weak	4
		1 hr.	5	weak	4
		2 hr.	5	weak	4
		4 hr.	5	weak	3
		8 hr.	5	weak	3
		16 hr.	5	weak	4
		24 hr.	5	weak	3
		continuous	5	medium	3
2	Nov. 8	7½ min.	5	medium	4
		15 min.	5	weak	5
		30 min.	5	weak	4
		1 hr.	5	weak	5
		2 hr.	5	weak	3
		4 hr.	5	weak	3
		8 hr.	5	weak	1
		16 hr.	5	medium	2
		24 hr.	5	medium	4
		continuous	5	weak	5

[1] Composition of ethylene-air mixture: 1.5 per cent ethylene and 98.5 per cent air.

by dilute solutions of copper. It is not at all clear why chemical stimulation by ethylene, copper solutions, etc., should induce the production of intumescences so similar to those reported by Sorauer, Küster, and others to arise in response to abnormal conditions of the physical environment, such as weak light, excessive humidity, and high temperature. Von Schrenk found, so far as he studied, that the formation of intumescences on cauliflower was not influenced by variations in these environmental conditions. I find that the production of intumescences on the stems of the apple in response to ethylene gas is little influenced by conditions of light intensity or of humidity. There is evidently no summation in these cases of chemical with light or moisture stimuli. Differences in temperature, on the other hand, have a marked effect on the number and

size of the intumescences formed by ethylene. The optimum appears from my results to be near 15° C., while the maximum seems to lie slightly above 30° C. According to Sorauer's view the opposite would be expected in case the ethylene and temperature stimuli do not antagonize each other.

The formation of intumescences in the bark and in the buds of woody stems seems to be one of the most pronounced responses to chemical stimulation so far reported for plants, viewed from the standpoint of the amount of the stimulus required to initiate the reaction. My results have shown even greater sensitiveness to ethylene than has been reported by other investigators.

Crocker and Knight (1908) found that flowers of the carnation close after exposure for twelve hours to 1 part of ethylene to 3,000,000 parts of air. The etiolated epicotyl of the sweet pea (Crocker, Knight, and Rose, 1913) was inhibited in its rate of growth when only 1 part of ethylene to 10,000,000 parts of air was present. E. M. Harvey (1913) reported definite droopings of the leaves and petioles of the castor bean plant in a similar concentration of ethylene, but mentioned that proliferation at the abscission layer failed to occur in concentrations lower than 1 part to 500,000 parts of air. It appears, therefore, that intumescence formation in the stems of Transparent apple is an even more delicate test for traces of this gas.

As described, I have obtained positive responses on cuttings of Transparent apple with 1 part of ethylene to 100,000,000 parts of air. My results indicate that the response in the distal ends of such cuttings may be more consistant and definite than those occurring in the buds and in the internodes. This stronger response at the distal end of a cutting may result from a more rapid penetration of the gas in this region. It is not clear that polarity has any influence on the response. Further experiments on these points are needed.

The fact that the entire range of concentrations of ethylene from extremely high to extremely low induce the formation of very similar outgrowths is in interesting contrast to what is regarded, in general, as the effect of so-called toxic agents on living organisms. Such substances retard growth or kill when used in large amounts, and many of them stimulate when used in small amounts. We have abundant evidence from the studies of Neljubow (1901), Crocker and Knight (1908), and

others that ethylene may be extremely toxic to some plants or plant parts, and may prove lethal when present in the merest traces.

That ethylene may hasten the disappearance of chlorophyll in some plants and plant parts has been well shown by the work of Denny (1924), Rose (1925b), and others. In view of this, it is all the more noteworthy that the chlorenchyma of stems of apple shows no decrease in the chlorophyll content when treated with this gas, in either high or low concentrations. Studies by E. M. Harvey (1913) and Denny (1923) indicate that ethylene increases the rate of plant metabolism. The disappearance of chlorophyll in materials treated with ethylene may not be a direct effect of the ethylene itself, but rather an indirect effect due to the resultant increased rates of the metabolic activities of the treated materials. All materials, such as the fruits of lemons and tomatoes and leaves of celery, which have shown an acceleration in the disappearance of chlorophyll when treated with ethylene are tissues from which the chlorophyll has already begun to disappear, or from which it soon would normally disappear. Since the chlorenchyma of stems would normally retain its green color, it is perhaps only natural to find that for it ethylene treatment does not result in bleaching.

SUMMARY

Extensive experiments made on cuttings of Transparent apple have shown that ethylene stimulation may induce the formation of intumescences under a wide range of environmental and biological conditions. Materials collected from different parts of the same individual or from different individuals of the same variety showed no apparent variation in the nature or extent of the reactions. Tests made during every month of the year indicate no pronounced seasonal or periodic variation in the sensitiveness of the tissues to ethylene stimulation. Humidities ranging from 67 per cent to 92 per cent have no apparent effect on the ethylene response. No correlation was apparent between the number or extent of the intumescences and the percentage of humidity.

Temperature exerts a marked influence on the number and size of the intumescences. The optimum temperature for their formation is near 15° C., while 30° C. is about the maximum

at which they will form. A much larger relative number of the cuttings responded at the optimum than at higher temperatures.

The intumescences form equally well in continuous light, continuous darkness, or in alternate light and darkness.

Ethylene and illuminating gas gave about the same responses when run in parallel series of concentrations from .05 to 50 per cent.

Intumescences do not form in the absence of oxygen. They do not develop in the presence of 25 per cent or more of carbon dioxide.

Mixtures varying all the way from 75 per cent of ethylene and 25 per cent of oxygen to 1 part of ethylene to 1,000,000 or 100,000,000 parts of air will induce the formation of intumescences at least in the ends of cuttings of Transparent apple. No definite differences in the extent or number of the responses were observed for concentrations ranging from 75 per cent ethylene and 25 per cent of oxygen to 1 part of ethylene to 10,000 parts of air. Intumescences formed in concentrations of ethylene of 1 part to 1,000,000 or 100,000,000 parts of air were much less extensive and were slower in appearing than those in the higher concentrations, but they were none-the-less distinct and characteristic.

Exposures to a mixture of 1.5 per cent ethylene and 98.5 per cent air for periods ranging from 7½ minutes to continuous exposure through the experimental period all gave responses. No variation was observed in the number or extent of the intumescences formed with this range of exposure times.

Ethylene appeared to have no bleaching effect on the chlorenchyma of the stems of Transparent apple.

I wish to express my appreciation to Professor R. A. Harper for stimulating criticism and invaluable assistance during the progress of these studies and in the preparation of this paper. I also greatly appreciate the Edna L. Smith fellowship in botany which made possible the completion of these studies.

COLUMBIA UNIVERSITY,
NEW YORK CITY.

Literature Cited

Atkinson, G. F. 1893a. Oedema of the tomato. Bull. Cornell Agr. Exp. Sta. 53: 1–108. pl. 1–8.

————. 1893b. Oedema of apple trees. Bull. Cornell Agr. Exp. Sta. 61: 299–302. fig. 1, 3.

Bayliss, J. S. 1907. On the galvantropism of roots. Ann. Bot. **21:** 387–405.

Crocker, W., and L. I. Knight. 1908. Effect of illuminating gas and ethylene upon flowering carnations. Bot. Gaz. **46:** 259–276.

————, **L. I. Knight, and R. C. Rose.** 1913. A delicate seedling test. Science **37:** 380–381.

Dale, E. 1901. Investigations on abnormal outgrowths or intumescences on *Hibiscus vitifolius*. Phil. Trans. Roy. Soc. London B **194:** 163.

————. 1899. Intumescences on *Hibiscus vitifolius*. Ann. Bot. **13:** 622–624.

Davis, H. V. 1910. Notes on certain intumescences in roots. New Phytologist **9:** 325–326.

Denny, F. E. 1924. Effect of ethylene upon respiration of lemons. Bot. Gaz. **77:** 322–329.

Doubt, S. L. 1917. The response of plants to illuminating gas. Bot. Gaz. **63:** 209–225. fig. 1–6.

Douglas, G. E. 1907. The formation of intumescences on potato plants. Bot. Gaz. **43:** 233–250.

Fujii, K. 1925. Ueber die Entlassung der Spermatozoiden von *Isoëtes*. Flora N. Ser. **18–19:** 115–126.

Haberlandt, G. 1899. Ueber experimentalle Hervorrufung eines neuen Organs bei *Conocephalus ovatus* Trec. Festchr. f. Schwendener, p. 104–119. fig. 1, 2.

Hahn, G. G., C. Hartley, and A. Rhoads. 1920. Hypertrophied lenticels on the roots of conifers and their relation to moisture and aeration. Jour. Agr. Res. **20:** 253–266.

Harvey, E. M. 1913. The castor bean plant and laboratory air. Bot. Gaz. **56:** 439–442.

————. 1915. Some effects of ethylene upon the metabolism of plants. Bot. Gaz. **60:** 193–214. fig. 1, 2.

————, **and R. C. Rose.** 1915. The effect of illuminating gas on root systems. Bot. Gaz. **60:** 27–44. fig. 1–8.

Harvey, R. B. 1925. Blanching of celery. Univ. Minnesota Agr. Exp. Sta. Bull. 222: 1–20. fig. 1–10.

Küster, E. 1903a. Pathologische Pflanzenanatomie.

————. 1903b. Ueber experimentelle erzeute Intumeszenzen. Ber. deutsch. Bot. Ges. **21:** 452–458.

Luckhardt, A. B., and J. B. Carter. 1923. Ethylene as a gas anaesthetic, preliminary communication. Jour. Am. Med. Assoc. **80:** 1440–1442.

Molz. 1906. Dropsy of the grape. Ber. der Kgl. Lehranstalt zu Geisenheim a. Rhein. (Cited by Sorauer.)

Norack, F. 1901. Eine Treibhauskrankheit der Weinreibe. Gartenflora **50:** 619–622

Nägeli. 1893. Oligodynamische Erscheinungen in lebenden Zellen. Denkschr. der Schweiz. Naturf. Ges. **33.**

Neljubow, D. 1901. Ueber die horizontale Nutation der Stengel von *Pisum sativum* und einiger anderen Pflanzen. Beih. Bot. Centralbl. **10:** 128–138.

Prillieux, Ed. 1892. Intumescences sur les feuilles d'oeillets malades. Bull. d. l. Soc. d. France **39:** 370.

Rose, J. T. 1925a. Shortening the rest period of potatoes with ethylene gas. Potato News Bull. **2:** 363–365.

————. 1925b. Physiological effects of ethylene on green mature tomato fruits. Absts. Plant Physiol. Sect. Bot. Soc. Amer., Kansas City, Missouri, 1925.

Rosen, H. R. 1916. The development of the *Phylloxera vasterix* leaf gall. Am. Jour. Bot. **3:** 337–360.

Rumbold, C. 1916. Pathological anatomy of the injected trunks of chestnut trees. Proc. Am. Phil. Soc. **55:** 485–493. pl. 15–18.

Schilling, E. 1915. Ueber hypertropische und hyperplastische Gewebeswucherungen an Sprossachsen, Versacht durch Paraffine. Jahrb. Wiss. Bot. **55:** 177–258. fig. 1–43.

Smith, E. F. 1917. Mechanism of tumor growth in crowngall. Jour. Agr. Res. **8:** 165–189. pl. 4–65.

Sorauer, P. 1886. Handbuch der Pflanzenkrankheiten. Auflage 1.

————. 1892. Nachweis der Verweichlichung unserer Obstbäume durch die Kulture. Zeit. f. Pflanzenkrankh. p. 66.

————. 1899. Ueber Intumeszenzen. Ber. deutsch. Bot. Ges. **17:** 456–460.

Steiner, R. 1905. Ueber Intumeszenzen bei *Rueillia formosa* und *Aphelandra Porteana*. Ber. deutsch. Bot. Ges. **23:** 105–113.

Stone, G. E. 1913. The effect of illuminating gas on vegetation. Ann. Rept. Massachusetts Exp. Sta. **25:** 45–60. fig. 1–3.

Taylor, W. R. 1919. On the production of new cell formations in plants. Contrib. Univ. Pennsylvania Bot. Lab. **4:** 271–299. pl. 71–78.

Trotter, A. 1904. Intumescenzes fogliari di *Ipomaea Batatas*. Annali di Botanica **1:** 365.

Viala, P., and P. Pacottet. 1904. Sur les verrues des feuilles de la vigne. Compt. Rend. de s. d. l'Acad. d. Sci. **138:** 163.

Von Schrenk, H. 1905. Intumescences formed as a result of chemical stimulation. Rept. Missouri Bot. Gard. **16:** 125–148. pl. 25–31.

Wallace, R. H. 1926. The production of intumescences upon apple twigs by ethylene gas. Bull. Torrey Bot. Club **53:** 385–401. pl. 14–15.

Wisniewski, P. 1910. Ueber Induktion von Lenticellenwucherungen bei *Ficus*. Bull. d. l'Acad. d. Sci. de Cracovoie. Ser. B Sci. Naturelles, no. 5, B, May, 1910. p. 359–367. 2 pl.

Wolf, F. A. 1918. Intumescences, with a note on mechanical injury as a cause for their formation. Jour. Agr. Res. **13:** 253–259.

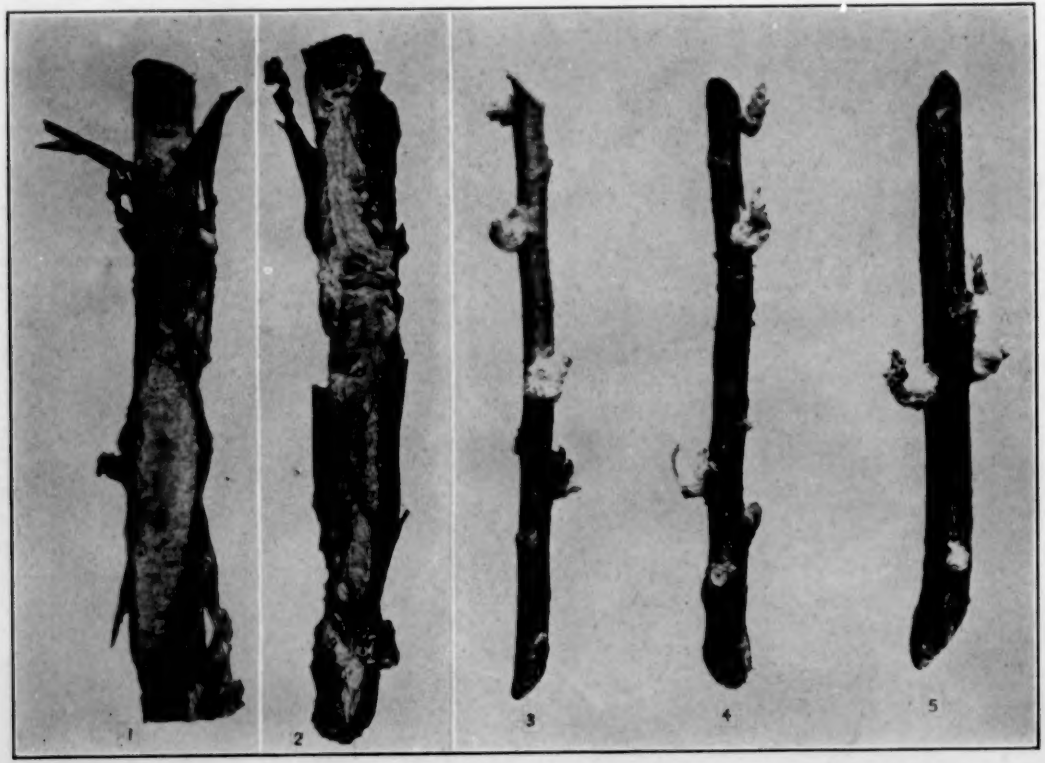
Explanation of Plate

PLATE 34

Fig. 1. Wide-spread but superficial intumescences on an internode of Transparent apple (× 1.5). The dead corky layers have been split and rolled back. The chlorenchyma and the deeper tissues are apparently normal.

Fig. 2. Wide-spread and deep-seated intumescences on an internode of Transparent apple (× 1.5). The chlorenchyma is somewhat exposed, but most of the response is localized in the deeper lying tissues.

Figs. 3, 4, and 5. Three cuttings of *Populus deltoides* (× 0.7) showing intumescences at the base of buds (one month after exposure to ethylene). Note that the tissue masses of the intumescences are more or less coherent and not loose and granular as they are in portions of the intumescences shown in figure 2.



INTUMESCENCES PRODUCED BY ETHYLENE

INDEX TO AMERICAN BOTANICAL LITERATURE
1925–1927

This Index is reprinted monthly on cards, and furnished in this form to subscribers at the rate of three cents for each card. Selections of cards are not permitted; each subscriber must take all cards published during the term of his subscription. Correspondence relating to the Index should be addressed to the Bibliographer of the Torrey Botanical Club, Mrs. Helen Smith Hill, Brooklyn Botanic Garden, Brooklyn, N. Y.

Aamodt, O. S. Breeding wheat for resistance to physiologic forms of stem rust. Jour. Am. Soc. Agron. **19**: 206–218. Mr 1927.

Anderson, H. W. The effect of sodium silicofluoride sprays on the peach and on the control of bacterial spot. Science II, **65**: 16–18. 7 Ja 1927.

Angst, L. The gametophyte of *Soranthera ulvoidea*. Publ. Puget Sound Biol. Sta. **5**: 159–162. *pl. 9*. D 1926.

Artschwager, E. Development of flowers and seed in the sugar beet. Jour. Agr. Res. **34**: 1–25. *f. 1–15* + *pl. 1*. 1 Ja 1927.

Atanasoff, D. Net necrosis of potato. Phytopathology **16**: 929–940. *f. 1–5*. 15 D 1926.

Bailey, D. L. Physiologic specialization in *Puccinia graminis avenae* Erikss. and Henn. Minnesota Agr. Exp. Sta. Tech. Bull. 35: 1–33. *pl. 1–7*. D 1925.

Bakke, A. L., Richey, H. W., and Reeves, K. Germination and storage of apple seeds. Iowa Agr. Exp. Sta. Res. Bull. 97: 243–255. *pl. 1, 2*. O 1926.

Bartram, E. B. Notes on *Mielichhoferia*. Bull. Torrey Club **54**: 31–34. *f. 1–20*. "Ja" 22 F 1927.

Bates, C. G. Better seeds, better trees. Jour. For. **25**: 130–144. *illust.* F 1927.

Belling, J. Multiple chromosomes and reduction division in flowering plants. Nature **119**: 122. 22 Ja 1927.

Berkeley, G. H. Studies in tomato streak. Sci. Agr **7**: 210–223. *f. 1–4.* F 1927.

B[erry], E. W. Frank Hall Knowlton. Science II, **65**: 7–8. 7 Ja 1927.
Born 2 S 1860. Died 22 N 1926.

Berry, E. W. Frank Hall Knowlton, 1860–1926. Am. Jour. Sci. V, **13**: 281–282. Mr 1927.
Born 2 S 1860. Died 22 N 1926.

Berry, E. W. The term Oligocene and certain climatic considerations. Am. Jour. Sci. V, **13**: 252–256. Mr 1927.

Black, J. M. *Olearia v. Shawia.* Jour. Bot. **65**: 17–18. Ja 1927.

Blake, S. F. New Asteraceae from Costa Rica. Jour. Washington Acad. Sci. **17**: 59–65. *illust.* 3 F 1927.
Two species in *Gnaphalium* and three in *Hieracium.*

Blakeslee, A. F. The chromosomal constitution of Nubbin, a compound (2n + 1) type in *Datura.* Proc. Natl. Acad. Sci. **13**: 79–85. *f. 1–4.* 15 F 1927.

Boyce, J. S. Observations on forest pathology in Great Britain and Denmark. Phytopathology **17**: 1–18. 15 Ja 1927.

Boyer, C. S. List of Quaternary and Tertiary Diatomaceae from deposits of southern Canada. Victoria Mem. Mus. Bull. Biol. Ser. 45: 1-26. 9 N 1926.

Breazeale, J. F. Alkali tolerance of plants considered as a phenomenon of adaptation. Arizona Agr. Exp. Sta. Tech. Bull. 11: 239–256. *f. 1–4.* 1 N 1926.

Britton, N. L. Studies of the Mimosas. Jour. N. Y. Bot. Gard. **28**: 67–68. Mr 1927.

Broadfoot, W. C. Studies on the parasitism of *Fusarium lini* Bolley. Phytopathology **16**: 951–978. *f. 1, 2.* 15 D 1926.

Brooks, M. M. Studies on the permeability of living cells, VII. The effects of light of different wave lengths on the penetration of 2,-6,-dibromo phenol indophenol into *Valonia.* Protoplasma **1**: 305–312. *f. 1, 2.* D 1926.

Brotherus, V. F. Contributions to the bryological flora of the Philippines, VI. Philipp. Jour. Sci. **31**: 277–300. N 1926.
Several species described as new.

Brown, H. B. The use of first generation crosses and other methods for improving southern varieties of corn. Mississippi Agr. Exp. Sta. Bull. 236: 1–31. *f. 1–3.* Jl 1926.

Brown, N. A. Sweet pea fasciation, a form of crowngall. Phytopathology **17**: 29–30. *pl. 1, 2.* 15 Ja 1927.

Burkholder, W. H. A new bacterial disease of the bean. Phytopathology **16**: 915–927. *f. 1, 2* + *pl.* 55. 15 D 1926.
Phytomonas medicaginis var. *phaseolicola.*

Christensen, J. J., and Stakman, E. C. Physiologic specialization and mutation in *Ustilago zeae.* Phytopathology **16**: 979–999. *f. 1–11.* 15 D 1926.

Chun, W. Y. New species and new combinations of Chinese plants. Jour. Arnold Arbor. **8**: 19-22. "Ja" 28 F 1927.
In *Ostrya, Cercis, Castanopsis, Lithocarpus, Vanieria, Phoebe, Notaphoebe, Benzoin, Actinodaphne,* and *Neolitsea.*

Clayton, E. E. Control of seedbed diseases of cruciferous crops on Long Island by the mercuric chloride treatment for cabbage maggot. N. Y. (Geneva) Agr. Exp. Sta. Bull. 537: 1–29. D 1926.

Clute, W. N. James H. Ferriss. Am. Fern Jour. **16**: 103–106. "D 1926" 2 F 1927.
Born 18 N 1849. Died 17 Mr 1926.

Coleman, D. A., Fellows, H. C., and Dixon, H. B. Testing wheat for protein, with a recommended method for making the test. U. S. Dept. Agr. Bull. 1460: 1–32. *f. 1–3.* D 1926.

Collings, G. H. The influence of boron on the growth of the soybean plant. Soil Sci. **23**: 83–104. *pl. 1.* F 1927.

Conklin, G. H. Collecting hepatics along Lake Superior in northern Minnesota. Bryologist **30**: 6-8. "Ja" Mr 1927.

Cook, M. T. Relationship of cane varieties to diseases. Jour. Dept. Agr. Porto Rico **9**: 277–281. O 1925.

Cook, O. F., and Hubbard, J. W. Primitive cottons in Mexico. Characters of native cottons including a wild species. Jour. Hered. **17**: 463–472. *f. 10–15.* "D 1926" 12 F 1927.

Coulter, J. M. The contribution of science to the welfare of the nation. Sci. Mo. **24**: 193–205. Mr 1927.

Dearness, J. *Amanita* poisoning. Mycologia **19**: 93–94. 1 Mr 1927.

Deatrick, E. P. Porous clay auto-irrigator cones for watering potted soil and plants. Jour. Am. Soc. Agron. **19**: 252. *f. 1, 2.* Mr 1927.

Dickey, L. B. The avocado and vitamines. Science II, **65**: 15. 7 Ja 1927.

Drayton, F. L., comp. A summary of the prevalence of plant diseases in the Dominion of Canada 1920–1924. Canada Dept. Agr. Bull. n. s. 71: 1–88. 1926.

Dungan, G. H. The influence of plant injury and the root rot diseases upon the physical and chemical composition of corn grain. Illinois Agr. Exp. Sta. Bull. 284: 253–281. *f. 1–9.* D 1926.

Durrell, L. W., and Lungren, E. A. Barberry eradication and sources of black stem rust in Colorado. Colorado Agr. Exp. Sta. Bull. 315: 1–18. *f. 1–10.* Ja 1927.

Emerson, R. The effect of certain respiratory inhibitors on the respiration of *Chlorella*. Jour. Gen. Physiol. **10**: 469–477. *f. 1–6.* 20 Mr 1927.

Essary, S. H. Two important tomato diseases and their control. Tennessee Agr. Exp. Sta. Circ. 8: 1–4. *illust.* Ja 1927.
Wilt and leaf-spot.

Evans, A. W., and Meyrowitz, R. Catalogue of the lichens of Connecticut. Connecticut Geol. & Nat. Hist. Surv. Bull. 37: 1–49. 1926.

Faris, J. A. Zonate food rot of sugar cane. Phytopathology **17**: 83–94. *f. 1–10.* 15 F 1927.

Faris, J. A., and Allison, R. V. Sugar cane root disease in Cuba: a progress report upon the root disease situation in 1925. Phytopathology **17**: 61–82. *f. 1–11.* 15 F 1927.

Farwell, O. A. Botanical gleanings in Michigan IV. Am. Midl. Nat. **10**: 199–219. Ja 1927.

Fassett, N. C. *Hieracium canadense* var. *hirtirameum* in northern Michigan. Rhodora **28**: 246. "D 1926" 28 Ja 1927.
Isle Royale.

Fassett, N. C. Notes on the flora of Boothbay, Maine—II. Rhodora **29**: 14–15. 28 Ja 1927.

Fernald, M. L. *Muhlenbergia uniflora.* Rhodora **29**: 10–14. 28 Ja 1927.
1 variety and 1 combination described as new.

Fernald, M. L. Two summers of botanizing in Newfoundland. Rhodora **28**: 49–63; 74–87; 89–111; 115–129; 145–155; 161–178; 181–204; 210–225; 234–241. *pl. 153–155.* 11 My; 11 Je; 22 Jl; 8 S; 20 S; 6 N; 20 D 1926; 28 Ja 1927.

Fisk, E. L. The chromosomes of *Zea mays.* Am. Jour. Bot. **14**: 53–75. *pl. 10.* "F" 8 Mr 1927.

Fleming, C. E., Miller, M. R., and Vawter, L. R. The common chokecherry (*Prunus demissa*) as a plant poisonous to sheep and cattle. Nevada Agr. Exp. Sta. Bull. 109: 1–30. *illust.* Mr 1926.

Flor, H. H. Fungicidal activity of furfural. Iowa State Coll. Jour. Sci. **1**: 199–223. Ja 1927.

Foote, Mrs. E. M. Ferns of Cornwall, Connecticut. Am. Fern Jour. **16**: 124–127. "D 1926" 2 F 1927.

Gager, C. S., and Blakeslee, A. F. Chromosome and gene mutations in *Datura* following exposure to radium rays. Proc. Natl. Acad. Sci. **13**: 75–79. 15 F 1927.

Gaines, E. F. The relation of triploid factors and chromosome groups in wheat and oats. Jour. Am. Soc. Agron. **19**: 202–205. Mr 1927.

Garber, R. J., and Quisenberry, K. S. The inheritance of length of style in buckwheat. Jour. Agr. Res. **34**: 181–183. 15 Ja 1927.

Garber, R. J., and Quisenberry, K. S. Natural crossing in oats at Morgantown, West Virginia. Jour. Am. Soc. Agron. **19**: 191–197. F 1927.

Garber, R. J., and Quisenberry, K. S. Self-fertilization in buckwheat. Jour. Agr. Res. **34**: 185–190. *f. 1.* 15 Ja 1927.

Gardner, N. L. New Rhodophyceae from the Pacific coast of North America. II. Univ. California Publ. Bot. **13**: 235–272. *pl. 24–35.* 28 F 1927.

Gill, T. The lifeblood of trees. Nat. Mag. **9**: 177–180. *illust.* Mr 1927.

Gleason, H. A. Four new Melastomataceae from Peru. Bull. Torrey Club **54**: 25–29. "Ja" 22 F 1927.
In *Axinaea, Tibouchina* and *Brachyotum.*

Gloyer, W. O. The dwarfing, shriveling, and dropping of cherries and prunes. New York (Geneva) Agr. Exp. Sta. Bull. 540: 1–18. *pl. 1–4.* D 1926.

Gravatt, A. Rathbun. A witches' broom of introduced Japanese cherry trees. Phytopathology **17**: 19–24. *f. 1.* 15 Ja 1927.

Grier, N. M. Mr. Taylor vs. "Grier's Notes on the Flora of Long Island." Rhodora **28**: 242–245. "D 1926" 28 Ja 1927.

Grier, N, M. A note on the interrupted fern. Torreya **27**: 8–9. F 1927.
Osmunda claytoniana.

Grout, A. J. Studies in *Ditrichum.* Bryologist **30**: 4–5. Ja 1927.
D. tortuloides described as new.

Haenseler, C. M. Plant diseases of New Jersey (twelfth instalment). Pea diseases. New Jersey Agr. **9**[2]: 6–7. F 1927.

Hahn, G. G. *Phomopsis juniperovora* and closely related strains

on conifers. Phytopathology **16:** 899–914. *f. 1* + *pl. 52–54.* 15 D 1926.

Harland, S. C., Haigh, J. C., and Lochrie, J. L. On a progressive variation with age of a simple Mendelian ratio in the cowpea. Genetica **8:** 507–512. 1926.

Harper, R. M. A six-hour cross-section of the vegetation of southern Ontario. Torreya **27:** 1–8. F 1927.

Harris, J. A. The cat tail, *Typha angustifolia*, in Utah. Torreya **27:** 9–11. F 1927.

Harris, T. M. The rhaetic flora of Scoresby Sound, East Greenland. Meddel. Grønland **68:** 45–147. *f. 1–34* + *pl. 1–13.* 1926.

Hartley, C. Notes on *Hibiscus* diseases in West Java. Phytopathology **17:** 25–27. 15 Ja 1927.

Hauman, L. Étude phytogéographique de la Patagonie. Bull. Soc. Roy. Bot. Belg. **58:** 105–179. *f. 1–7* + *pl. 1–13.* 1926.

Henderson, N. F. Ohio mosses, Bryales. Ohio Jour. Sci. **27:** 1–18. "Ja" 15 F 1927.

Holm, T. The flower of *Chimaphila*. Rhodora **29:** 1–6. *pl. 156.* 28 Ja 1927.

Holm, T. Sciaphilous plant-types. Beihefte Bot. Centralbl. Abt. I, **44:** 1–89. *f. 1–3.* Ja 1927.

Hope, A. The dissemination of weed seeds by irrigation water in Alberta. Sci. Agr. **7:** 268–276. Mr 1927.

Humbert, E. P., and Mogford, J. S. Variation in certain lint characters in a cotton plant and its progeny. Texas Agr. Exp. Sta. Bull. 349: 1–23. *f. 1–4.* F 1927.

Hunt, W. R. The Uredinales or rusts of Connecticut and the other New England states. Connecticut Geol. & Nat. Hist. Surv. Bull. 36: 1–198. 1926.

Hurd-Karrer, A. M., and Hasselbring, H. Effect of smut (Ustilago zeae) on the sugar content of cornstalks. Jour. Agr. Res. **34:** 191–195. 15 Ja 1927.

Jackson, A. B. The *Fusarium* wilt of China asters. Sci. Agr. **7**: 233–247. *pl. 1–3.* Mr 1927.

Jackson, H. S. The rusts of South America based on the Holway collections—II. Mycologia **19**: 51–65. 1 Mr 1927.
Many species described as new.

Jagger, I. C. Powdery mildew of muskmelons in the Imperial Valley of California in 1925. Phytopathology **16**: 1009–1010. 15 D 1926.

Jennings, O. E. Meanings of some plant names. Bull. Gard. Club Am. 13: 16–19. Ja 1927.

Johnston, C. O. Effects of soil moisture and temperature and of dehulling on the infection of oats by loose and covered smuts. Phytopathology **17**: 31–36. 15 Ja 1927.

Johnston, I. M. Studies in the Boraginaceae—VI. A revision of the South American Boraginoideae. Contr. Gray Herb. 78: 1–118. 15 Mr 1927.
Many species described as new.

Kern, F. D. A trip to Santo Domingo. Sci. Mo. **23**: 529–538. D 1926.

Kern, F. D., and Whetzel, H. H. Hemibasidiomycetes. (in Seaver, F. J., & Chardon, C. E. Botany of Porto Rico and the Virgin Islands. Mycology. Sci. Survey Porto Rico, 8: 107–144. 29 D 1926.)

Keyes, F. J. *Trillium grandiflorum* in Maine. Rhodora **29**: 15. 28 Ja 1927.
At Chesterville.

Kidder, A. V. Eskimos and plants. Proc. Natl. Acad. Sci. **13**: 74–75. 15 F 1927.

Kirk, L. E. Breeding improved varieties of forage crops. Jour. Am. Soc. Agron. **19**: 225–239. Mr 1927.

Kirkwood, J. E. Botanical exploration in the Rocky Mountains. Sci. Mo. **24**: 236–250. Mr 1927.

Kittredge, E. M. A remarkable form of walking fern. Am. Fern. Jour. **16**: 98–100. *pl. 8.* 28 S 1926.

Lantz, C. W. Respiration in corn with special reference to catalase. Am. Jour. Bot. **14**: 85–105. *pl. 1–4.* "F" 8 Mr 1927.

LaRue, C. D. The Hevea rubber tree in the Amazon Valley. U. S. Dept. Agr. Bull. 1422: 1–69. O 1926.

Leighty, C. E. Breeding plants for disease resistance. Jour. Am. Soc. Agron. **19**: 219–225. Mr 1927.

Leonard, E. C. The North American species of *Scutellaria*. U. S. Natl. Herb. Contr. **22**: i–vii, 703–748. 1927.

Letcher, H., and Willaman, J. J. Biochemistry of plant diseases. VIII. Alcoholic fermentation of *Fusarium lini*. Phytopathology **16**: 941–949. *f. 1–3.* 15 D 1926.

Lillibridge, A. E. Some abnormal forms of *Botrychium simplex*. Am. Fern Jour. **16**: 127–128. *pl. 11.* "D 1926" 2 F 1927.

Lindstrom, E. W., and Gerhardt, F. Inheritance of carbohydrates and fat in crosses of Dent and sweet corn. Iowa Agr. Exp. Sta. Res. Bull. 98: 259–277. N 1926.

Linford, M. B. Additional hosts of *Aphanomyces euteiches*, the pea rootrot fungus. Phytopathology **17**: 133–134. 15 F 1927.

Lute, A. M. Alfalfa seed made permeable by heat. Science II, **65**: 166. 11 F 1927.

Lutz, B. Estudos sobre a biologia floral de *Mangifera indica* L. Arch. Mus. Nac. Rio. de Janeiro **26**: 125–158. *pl. 1–4.* 1926.

MacGillivray, J. H. Effect of phosphorus on the composition of the tomato plant. Jour. Agr. Res. **34**: 97–127. *f. 1–6.* 15 Ja 1927.

McHargue, J. S. Significance of the occurrence of manganese, copper, zinc, nickel and cobalt in Kentucky blue grass. Ind. and Eng. Chem. **19**: 273–276. *illust.* F 1927.

Mackenzie, K. K. Further light on our purple-flowered Eupatoriums. Rhodora **29**: 6–9. 28 Ja 1927.

Mackenzie, K. K. *Solidago conferta* Miller. Rhodora **29:** 17–19. "F" 28 Ja 1927.

Mackenzie, K. K. Technical name of sugar maple. Rhodora **28:** 233–234. "D 1926" 28 Ja 1927.

Mackenzie, K. K. Usage. Rhodora **29:** 26–32. "F" 28 Ja 1927.

MacMillan, H. G. A method of growing bacteriologically sterile potato plants. U. S. Dept. Agr. Bull. 1465: 1–21. *f. 1–6 + pl. 1–6.* Ja 1927.

McNair, J. B. Citrus products. I. Publ. Field Mus. Nat. Hist. Bot. Ser. **6:** 1–212. *pl. 1–6.* Au 1926.

Mangels, C. E. A study of dark, hard kernel and protein content of hard red spring wheat. Jour. Agr. Res. **34:** 157–166. *f. 1–6.* 15 Ja 1927.

Mangelsdorf, P. C. Progress and possibilities in forage crop improvement. Jour. Am. Soc. Agron. **19:** 239–242. Mr 1927.

Markgraf, F. Neue Apocynaceen aus Südamerika. II. Notizbl. Bot. Gart. Berlin **9:** 959–963. 15 N 1926.

Mattoon, W. R. Loblolly pine primer. U. S. Dept. Agr. Farmers Bull. 1517: 1–38. *f. 1–27.* D 1926.

Meisel, M. A bibliography of American natural history. The pioneer century, 1769–1865. Vol. II. The institutions which have contributed to the rise and progress of American natural history, which were founded or organized between 1769 and 1844. 1-741. 1927.

Melander, L. W., and Craigie, J. H. Nature resistance of *Berberis* spp. to *Puccinia graminis.* Phytopathology **17:** 95–114. *f. 1–4.* 15 F 1927.

Melchior, H. Plantae Steinbachianae. Botizbl. Bot. Gart. Berlin **9:** 1034–1043. 15 N 1926.
Several species described as new.

Mendiola, N. B. Imperfect hermaphroditism in flowers of *Hibiscus,* removed by surgical operation. Philipp. Jour. Sci. **32:** 65–74. *pl. 1–4.* Ja 1927.

Merrill, E. D. New Chinese ligneous plants. Jour. Arnold Arbor. **8**: 3–19. "Ja" 28 F 1927.

Two genera, *Phoebe* and *Bennettiodendron* as well as many species, described as new.

Milbraed, J. Plantae tessmannianae peruvianae III. Notizbl. Bot. Gart. Berlin **9**: 964–977. 15 N 1926.

2 genera, *Tessmanniacanthus* and *Aphelandrella*, as well as many species, described as new.

Mowry, H. Palms of Florida. Florida Agr. Exp. Sta. Bull. 184: 1–54. *f.* 1–57. O 1926.

Moxley, E. A. Some November notes from Owen Sound, Ontario. Bryologist **30**: 8. "Ja" Mr 1927.

Murneek, A. E. Physiology of reproduction in horticultural plants. I. Reproduction and metabolic efficiency in the tomato. Missouri Agr. Exp. Sta. Res. Bull. 90: 1–19. S 1926.

Murrill, W. A. The edibility of *Leotia*. Mycologia **19**: 92. 1 Mr 1927.

Murrill, W. A. *Spongipellis fissilis*. Mycologia **19**: 90–91. 1 Mr 1927.

Navez, A. E. "Galvanotropism" of roots. Jour. Gen. Physiol. **10**: 551–558. *f.* 1. 20 Mr 1927.

Nelson, C. I., and Dworak, M. Studies on the nature of wilt resistance in flax. North Dakota Agr. Exp. Sta. Bull. 202: 1–30. *f.* 1. S 1926.

Nolla, J. A. B. A new *Alternaria* disease of onions (*Allium cepa* L.) Phytopathology **17**: 115–132. *pl.* 3–5. 15 F 1927.

A. allii.

Overholts, L. O. Eubasidiomycetes. (in Seaver, F. J., & Chardon, C. E. Botany of Porto Rico and the Virgin Islands. Mycology. Sci. Survey Porto Rico **8**: 145–181.) 29 D 1926.

Pack, A. N. Garden nature. Nat. Mag. **9**: 167–170. [8] *col. pl.* Mr 1927.

Palmer, E. J. On Nuttall's trail through Arkansas. Jour. Arnold Arbor. **8**: 24–55. "Ja" 28 F 1927.
Quercus Nuttallii described as new.

Platz, G. A., Durrell, L. W., and Howe, M. F. Effect of carbon dioxide upon the germination of chlamydo-spores of *Ustilago zeae* (Beckm.). Ung. Jour. Agr. Res. **34**: 137–147. *f. 1–3.* 15 Ja 1927.

Plitt, C. C. Succession in lichens. Bryologist **30**: 1–4. "Ja" Mr 1927.

Pretz, H. W. On *Eragrostis peregrina* and its relatives. Rhodora **29**: 19–26. "F" 28 Ja 1927.

Randolph, F. R. A cytological study of two types of variegate pericarp in maize. New York (Cornell) Agr. Exp. Sta. Memoir 102: 1–14. *f. 1–4 + pl. 1, 2.* Au 1926.

Record, S. J. A new species of *Mollia* of British Guiana. Yale Univ. School For. Tropical Woods 9: 8–10. 1 Mr 1927.
M. sphaerocarpa.

Record, S. J. Occurrence of "ripple marks" in woods. Yale Univ. School For. Tropical Woods 9: 13–18. 1 Mr 1927.

Reed, H. S. Growth and differentiation in plants. Quar. Rev. Biol. **2**: 79–101. *f. 1.* Mr 1927.

Rehder, A. *Pottsia grandiflora.* Jour. Arnold Arbor. **8**: 68. "Ja" 28 F 1927.

Rehder, A. *Pseudocytisus* and *Vella.* Jour. Arnold Arbor. **8**: 22–24. "Ja" 28 F 1927.

Rehder, A. The varietal categories in botanical nomenclature and their historical development. Jour. Arnold Arbor. **8**: 56–68. "Ja" 28 F 1927.

Robinson, B. L. [*Eupatorium* and *Ophryosporus*]. (in Standley, P. C. Trees and shrubs of Mexico. Contr. U. S. Natl. Herb. **23**: 1432–1470. 15 N 1926.)

Rodenhiser, H. A. Physiologic specialization of *Ustilago nuda* and *Ustilago tritici.* Phytopathology **16**: 1001–1007. *f. 1.* 15 D 1926.

Rydberg, P. A. Notes on Fabaceae—VIII. *Hamosa.* Bull. Torrey Club **54**: 13–23. "Ja" 22 F 1927.
Several species described as new.

Sande-Bakhuyzen, H. L. van de, and Alsberg, C. L. The growth curve in annual plants. Physiol. Rev. **7**: 151–187. Ja 1927.

Scarth, G. W. The influence of external osmotic pressure and of disturbance of the cell surface on the permeability of *Spirogyra* and acid dyes. Protoplasma **1**: 204–213. S 1926.

Schaffner, J. H. Sex-limited characters in heterosporous sporophytes. Ohio Jour. Sci. **27**: 19–24. "Ja" 15 F. 1927.

Scott, F. M. On certain fossil cones from the Pacific coast. Bull. Torrey Club **54**: 7–11. "Ja" 22 F 1927.

Scott, I. T. Some protein analogies of the mycelium of *Fusarium Lycopersici.* Missouri Agr. Exp. Sta. Res. Bull. 92: 1–44. *f. 1–9.* O 1926.

Seaver, F. J. A tentative scheme for the treatment of the genera of the Pezizaceae. Mycologia **19**: 86–89. 1 Mr 1927.

Seaver, F. J., and Chardon, C. E. Botany of Porto Rico and the Virgin Islands. Mycology. Sci. Survey Porto Rico **8**[1]: 1–208. 29 D 1926.
Contributions by R. A. Toro, F. D. Kern, H. H. Whetzel and L. O. Overholts.

Seaver, F. J., Whetzel, H. H., and Westcott, C. Studies on Bermuda fungi. I. *Poronia leporina.* Mycologia **19**: 43–50. *f. 1–5* + *pl. 1–5.* 1 Mr 1927.

Seifriz, W. The physical properties of erythrocytes. Protoplasma **1**: 345–365. *f. 1–7* + *pl. 6, 7.* D 1926.

Serrano, F. B. Deterioration of abaca (Manila hemp) fiber through mold action. Philipp. Jour. Sci. **32**: 75–101. *f. 1, 2* + *pl. 1–10.* Ja 1927.

Setchell, W. A. *Zostera marina latifolia:* ecad or ecotype? Bull. Torrey Club **54**: 1–6. "Ja" 22 F 1927.

Shaner, R. F. Lamarck and the evolution theory. Sci. Mo. **24:** 251–255. Mr 1927.

Shear, C. L. Mycology in relation to human pathology. Am. Nat. **61:** 151–159. Mr–Ap 1927.

Shull, G. H. A heterozygous phenotype in shepherd's purse. Hereditas **9:** 225–235. *f. 1–6.* Ja 1927.

Small, J. K. Among floral aborigines. A record of exploration in Florida in the winter of 1922. Jour. N. Y. Bot. Gard. **28:** 1–20; 25–40. *f. 1–5.* Ja, F 1927.

Sorokin, H. A study of meiosis in *Ranunculus acris.* Am. Jour. Bot. **14:** 76–84. *pl. 13–15.* "F" 8 Mr 1927.

Spaeth, C. P., and Kraybill, H. R. A biochemical study of the false-blossom disease of the cranberry. Jour. Agr. Res. **34:** 35–47. 1 Ja 1927.

Sprague, G. F. Heritable characters of maize. XXVII.—Colored scutellum. Jour. Hered. **18:** 41–44. *f. 16.* "Ja" 9 Mr 1927.

Sprague, T. A. *Alibertia* or *Cordiera.* Jour. Bot. **65:** *16–17.* Ja 1927.

Sprague, T. A. *Cerefolium* Haller. Jour. Bot. **65:** 15–16. Ja 1927.

Standley, P. C. A counterfeit collection of Mexican plants falsely attributed to Brother G. Arsene. Science II, **65:** 130–133. 4 F 1927.

Standley, P. C. Poisonous trees of Central America. Yale Univ. School For. Trop. Woods 9: 3–7. 1 Mr 1927.

Standley, P. C. *Primula Parryi.* Gard. Chron. III, **81:** 27. *f. 1.* 8 Ja 1927.

Standley, P. C. Three new species of plants collected in British Honduras by Harry W. Winzerling. Yale Univ. School For. Trop. Woods 9: 10–12. 1 Mr 1927.

Hyperbaena Winzerlingii, Banisteria heterocarpa and *Lycianthes hypo leuca.*

Stebbins, G. L. Two plants new to Mt. Katahdin. Rhodora **29**: 15–16. 28 Ja 1927.
Epilobium lactiflorum and *Draba fladnizensis.*

Stewart, G., and Tingey, D. C. A method for controlling pollination of sugar beets. Jour. Am. Soc. Agron. **19**: 126–127. F 1927.

Stout, A. B., McKee, R. H., and Schreiner, E. J. The breeding of forest trees for pulp wood. Jour. N. Y. Bot. Gard. **28**: 49–63. *f. 1–7.* Mr 1927.

Taylor, A. M. Moss habitats of the western Catskills. Bryologist **30**: 9–12. "Ja" Mr 1927.

Toro, R. A. Fungi of Santo Domingo. I. Mycologia **19**: 66–85. *pl. 6.* 1 Mr 1927.
Several species described as new.

Toro, R. A. Pseudoperisporiales (in Seaver, F. J., and Chardon, C. E. Botany of Porto Rico and the Virgin Islands. Mycology. Sci. Surv. Porto Rico. **8**: 40–41. 29 D 1926).

Twining, F. S. A western currant. Nat. Mag. **9**: 151. *illust.* Mr 1927.
Ribes glutinosum.

Wainio, E. O. Lichenes Mexicani a F.M. Liebmann annis 1841–1843 collecti, in Museo Hauniensi asservati. Dansk Bot. Arkiv. 4^{11}: 1–25. 10 F 1926.

Waldron, L. R. Hybrid selections of Marquis and Kota, a comparative study with regard to disease resistance, yield and baking quality. No. Dakota Agr. Exp. Sta. Bull. 200: 1–64. *f. 1–11.* S 1926.

Walker, J. C. Diseases of cabbage and related plants. U. S. Dept. Agr. Farmers Bull. 1439: 1–30. *f. 1–12.* F 1927.

Walker, M. N., and Stahl, C. F. Certain grass hosts of the sugar cane mosaic disease and of the corn aphid considered in relation to their occurrence in Cuba. Trop. Pl. Res. Foundation Bull. 5: 1–14. 1926.

Weatherby, C. A. *Equisetum pratense* in Berkshire County, Massachusetts. Rhodora **29**: 32. "F" 28 Ja 1927.
New Marlboro.

Weatherby, C. A. Imperfectly circinate vernation in ferns. Am. Fern. Jour. **16**: 109–111. *f. 1–3.* "D 1926" 2 F 1927.

Weber, G. F., and Ramsey, G. B. Tomato diseases in Florida. Florida Agr. Exp. Sta. Bull. 185: 61–138. *f. 58–99.* D 1926.

Weimer, J. L. Crown wart of alfalfa in the south. Phytopathology **16**: 1012. 15 D 1926.

Wherry, E. T. Observations on the woolly lipfern. Am. Fern Jour. **16**: 107–109. "D 1926" 2 F 1927.

Whetzel, H. H., and Melhus, I. E. Joseph Rosenbaum. 1887–1925. Phytopathology **16**: 895–897. *port.* 15 D 1926.

Wilson, C. L. Adventitious roots and shoots in an introduced weed. Bull. Torrey Club **54**: 35–38. *f. 1, 2.* "Ja" 22 F 1927.
Roripa austriaca

Wilson, E. H. *Juniperus procera* Hochst. Jour. Arnold Arbor. **8**: 1–2. *pl. 4.* "Ja" 28 F 1927.

Wilson, H. K., and Hottes, C. F. Wheat germination studies with particular reference to temperature and moisture relationships. Jour. Am. Soc. Agron. **19**: 181–190. *f. 1.* F 1927.

Wingard, S. A. Black end of apple. Phytopathology **16**: 1011–1012. *f. 1.* 15 D 1926.

Wylie, R. B. Leaf structure and wound response. Science II, **65**: 47–50. 21 Ja 1927.